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Patent application No. Demande de brevet n° Patentanmeldung Nr.

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PRIORITY DOCUMENT



Der Präsident des Europäischen Patentamts; Im Auftrag

For the President of the European Patent Office

Le Président de l'Office européen des brevets p.o.

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Polypeptides capable of forming antigen binding structures with specificity for the Rhesus D antigens, the DNA encoding them and the process for their preparation and use

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Polypeptides capable of forming antigen binding structures with specificity for the Rhesus D antigens, the DNA encoding them and the process for their preparation and use

This invention relates to polypeptides forming antigen binding structures with specificity for Rhesus D antigens and especially to Fab molecules with specificity for the Rhesus D antigen. The invention also relates to their application to provide pharmacological and diagnostic compositions. The above Fab fragments when genetically engineered to be part of complete antibodies are useful for the prophylaxis of hemolytic disease of the newborn (HDN). This invention provides the novel DNA and amino acid sequences of the above polypeptides.

Thus, the antibodies can be used for the protection of Rhesus negative women before or immediately after the birth of a Rhesus positive child to prevent HDN in subsequent pregnancies.

The invention also includes the application of the said Fab molecules either alone or in combination with Fc constant regions as complete antibodies for the purposes of treating other illnesses which might benefit from anti-Rhesus D immunoglobulin e.g. treatment of idiopathic thrombocytopenic purpura (ITP).

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In addition anti-Rhesus D immunoglobulin can be used after mistransfusions of Rhesus positive blood to Rhesus negative recipients in order to prevent sensitization to the Rhesus D antigen. Further the invention relates to the application of these Fab fragments and antibodies as diagnostic reagents.

HDN is the general designation for hemolytic anemia of fetuses and newborn babies caused by antibodies of the mother. These antibodies are directed against antigens on the surface of the fetal erythrocytes. These antigens can belong to the Rhesus, ABO or other blood group systems.

The Rhesus blood group system includes 5 major antigens: D, C, c, E and e (Issitt, P.D., Med. Lab. Sci. 45:395, 1988). The D antigen is the most important of these antigens as it is highly immunogenic eliciting anti-Rhesus D antibodies during Rhesus incompatible pregnancies and following transfusion of Rhesus incompatible blood. The D antigen is found in approximately 85% of Caucasians in Europe and those individuals are said to be Rhesus positive. Individuals lacking the D antigen are called Rhesus negative. The expression of the D antigen can vary due to either low antigen density, hereafter known as weak D or D^u, or due to partial antigenicity, hereafter known as partial D antigens.

The Rhesus D antigen, a membrane protein of the erythrocyte, has recently been cloned and its primary structure described (Le Van Kim, C., et al., PNAS 89:10925, 1992). Modeling studies suggest that the Rhesus D antigen has 12 transmembrane domains with only very short connecting regions extending outside the cell membrane or protruding into the cytoplasm.

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The partial D phenotypes were first identified in people who carried D antigen on their red cells but who had an alloanti-D in their sera (Rose, R. R. and Sanger, R., Blood groups in man, Blackwell Scientific, Oxford, U.K. 1975; Tippett, P. et al., Vox Sanguinis. 70:123, 1996). This can be explained by regarding the D antigen as a mosaic structure with at least 9 different epitopes (epD1 to epD9). Thus in some D variant people the red cells lack part of this mosaic and antibodies are made to the missing D epitopes. Rhesus positive individuals that make antibodies against partial D antigens have been classified into six main different categories (D^{\parallel} to $D^{\vee \parallel}$) each having a different abnormality in the D antigen. More recently it has been shown that these D categories gave different patterns of reaction when tested against panels of human monoclonal anti-D antibodies (Tippett, P., et al., Vox Sanguinis. 70:123, 1996). The different reaction patterns identified the 9 epitopes and so define the different partial D categories. The number of epitopes present on the D antigen varies from one partial D category to another with the D^{VI} category expressing the least, epD3, 4 and 9. The D^{VI} category is clinically important as a D^{VI} woman can be immunized strongly enough to cause hemolytic disease of the newborn.

The prophylactic efficacy of anti-RhD IgG for prevention of hemolytic disease of the newborn is well established and has been in routine use for many years. As a result this severe disease has become a rarity. Nevertheless the underlying cause of the disease, i.e. RhD incompatibility between a RhD negative mother carrying a RhD positive child still remains and thus requires a continual supply of therapeutic anti-RhD IgG.

In recent years the assurance of a continual supply of anti-RhD IgG has become an increasing problem. The pool of available hyperimmune serum from alloimmunized multiparous Rhesus negative women has drastically decreased due to the success of prophylactic anti-RhD. Thus the current methods of production require repeated immunization of an increasingly reluctant pool of donors for the production of high titer antiserum (Selinger, M., Br. J. Obstet. Gynaecol. 98:509, 1991). There are also associated risk factors and technical problems such as the use of Rhesus positive red blood cells for repeated immunization carrying the risk of transmission of viral diseases like hepatitis B, AIDS and other as yet unknown viruses (Hughes-Jones, N.C., Br. J. Haematol. 70:263, 1988). Therefore an alternative method for production of anti-RhD antibodies is required.

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In the past few years various alternative sources of hyperimmune serum have been tried but all are associated with disadvantages. Epstein Barr Virus (EBV) transformation of lymphocytes creating B lymphoblastoid cell lines that secrete specific antibody including against the Rhesus D antigen (Crawford et al., Lancet. 386 Feb. 19th, 1983) are unstable and require extensive cloning. Also due to the low transformation efficiencies (1-3% of B cells) only a restricted range of antibody specificities can be obtained from the potential repertoire. Additionally it seems that mice do not respond to the Rhesus D antigen and thus no murine monoclonal antibodies are available which could be used for producing chimaeric or humanised antibodies. Until recently the only other alternative was production of human antibodies by the hybridoma technique which was also restricted by the lack of a suitable human myeloma cell fusion partner (Kozbor, D. and Roder, J.C., Immunol. Today. 4:72, 1983).

It is thus the object of the present invention to provide Fab fragments having a reactivity against the Rhesus D antigen as well as complete antibodies comprising the Fab fragments which are free from the above mentioned drawbacks.

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In the last few years the technique of repertoire cloning and the construction of phage display libraries has opened up new possibilities to produce human antibodies of defined specificity (Williamson, R.A. et al., PNAS 90:4141, 1993). These methods were thus applied to the preparation of polypeptides capable of forming antigen binding structures with specificity for Rhesus D antigens, especially of Fab fragments having an activity against Rhesus D and partial D antigens.

The generation of human antibodies by repertoire cloning as described in recent years (Barbas III, C.F. and Lerner, R.A., Companion Methods Enzymol. 2:119, 1991) is based on isolating mRNA from peripheral B cells. This method offers the tools to isolate natural antibodies, autoantibodies or antibodies generated during the course of an immune response (Zebedee, S.L., et al., PNAS 89:3175, 1992; Vogel, M. et al., Eur.J. Immunol. 24:1200, 1994). This method relies on constructing a recombinant antibody library from a particular donor starting from the mRNA coding for immunoglobulin (Ig) molecules. As only the peripheral blood lymphocytes (PBL) can be isolated from a donor the chances of finding specific antibody producing B cells in the periphery are increased if an individual is boosted with the desired antigen shortly before harvesting the PBL (Persson, M.A.A., et al., PNAS 88:2432, 1991). The total RNA is then isolated and the mRNA of the Ig repertoire can be cloned using Ig specific primers in the polymerase chain reaction (PCR) followed by the co-expression of heavy and light chains of the Ig molecule on the surface of a filamentous phage particle thereby forming an "organism" that in analogy to a B cell can bind to an antigen. In the literature this method is also known as the combinatorial approach as it allows the independent combining of heavy and light chains to form a functional Fab antibody fragment attached to one of the tail proteins, called pIII, of a filamentous phage. Phages carrying the Fab molecules (hereafter known as Phab particles) are selected for the desired antigen specificity, by a

5 process known as bio-panning. The antigen can be applied to a solid support, specific Phab bind to the antigen whilst non specific Phab are washed away and finally the specific Phab are eluted from the solid support. The specific Phab are then amplified in bacteria, allowed to re-bind to the antigen on the solid support and the whole process of bio-panning is repeated. The successive rounds of panning and amplification of selected Phab in bacteria result in an enrichment of specific Phab that can be seen from a rise in titer of colony forming units (cfu) plated out after each round of panning. Our previous experience and published data indicate that specific phage can usually be detected after 4 to 6 panning rounds (Vogel, M. et al., Eur.J. Immunol. 24:1200, 1994). In the above cited related art there is , however, no hint that the indicated steps can be used for a successful preparation of Fab fragments of anti-Rh D antibodies. In the appended figures 1a to 17b; DNA sequences coding for variable regions (V regions) of anti Rh D Fab fragments and the corresponding polypeptide sequences are disclosed. Fig. 18 shows the pComb3 expression system used according to the present invention. Figs. 19 and 20 show the separate preparation of genes of the heavy and light chains of the complete antibody according to the description ²20 in example 6. Subjects of the present invention are polypeptides capable of forming antigen binding structures with specificity for Rhesus D antigens according to the definition of claim 1. The table in claim 1 refers to the appended figures. The identification number for each sequence is given. The locations of the Rhesus D specific CDR1 (complementarity determining region 1), CDR2 and CDR3 regions are indicated in the figures and according to base pair number in the table of claim 1. Prefered polypeptides according to the invention are anti-Rhesus D antibodies which include the variable regions of the heavy and light chains according to the sequences given in 80619.DOC

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Figs. 1a -17b. The Figs. 1a, 2a, ... 17a are related to the variable regions of the heavy chain and the Figs. 1b, 2b, ... 17b are related to the variable regions of the light chain.

Further subjects of the present invention are the DNA sequences coding for antigen binding polypeptides according to the definition of claim 5. Prefered DNA sequences are those coding for variable regions of Fab fragments of anti-Rh D antibodies according to the Figs. 1a -17b. The Figs. 1a, 2a, ... 17a are related to the heavy chain and the Figs. 1b, 2b, ... 17b are related to the light chain.

A further subject of the present invention is the process for preparing recombinant Fab polypeptides according to the definition in claim 9.

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Further subjects of the present invention are anti-Rh D antibodies according to the definition of claim 10, preferably anti-Rh D immunoglobulin molecules comprising the heavy and light chain variable regions according to the Figs. 1a to 17b combined with known heavy and light chain constant regions.

Further subjects of the present invention are pharmaceutical and diagnostic compositions comprising polypeptides, anti-Rh D antibodies or Fab fragments according to the invention.

The total re-amplified Phab population obtained after each panning can be tested for specificity using various methods such as ELISA and immunodot assays. It is also defined by the nature of the antigen e.g. anti-Rhesus D Phabs are detected by indirect haemagglutination using a rabbit anti-phage antibody or equivalent Coombs reagent as the cross linking antibody. Once a total Phab population has been identified as positive for the desired antigen, individual Phab clones are isolated and the DNA coding for the desired Fab molecules is sequenced. Individual Fab can then be produced by use of the pComb3 expression system which is illustrated in Fig. 18. In this system the gIII gene, coding for the tail protein pIII, is cut out from the phagemid vector pComb3. This allows production of soluble Fab in the

bacterial periplasm. Such individual Fab fragments can then be tested for antigen specificity.

The phage display approach has also been used as a means of rescuing monoclonal antibodies from unstable hybridoma cell lines. This has been reported for anti-Rhesus D antibodies (Siegel, D.L. and Silberstein, L.E., Blood. 83:2334, 1994; Dziegiel, M. et al., J. Immunol. Methods. 182:7, 1995). A phage display library constructed from non-immunized donors has also been used to select Fv fragments (i.e. variable regions of heavy and light chains, V_H and V_L) specific for human blood group antigens which included one Fv fragment reacting against the Rhesus D antigen (Marks, J.D. et al., Biotechnology. 11:1145, 1993).

Important considerations when constructing combinatorial libraries are the source of cells used for RNA extraction and the nature of the antigen used for panning. Therefore, this invention uses a hyperimmune donor who was boosted i.v. with Rhesus D⁺ red blood cells (rbc). The PBL of the donor were harvested at +5 and +18 days after the i.v. boost and were used to construct 2 combinatorial libraries hereafter known as library D1 (LD1) and library D2 (LD2) respectively. Double immunofluorescence analysis of the harvested PBL, using the markers CD20 and CD38 for pan B cells and lymphoblastoid cells respectively, showed a higher than normal percentage of lymphoblastoid B cells, of plasma cell morphology. The high number of plasma cells found in the peripheral blood is most unusual as normally there are less than 1% in the periphery and probably indicates that the donor had a high percentage of circulating B cells with specificity for the Rhesus D antigen.

After construction of the library, the selection of Phabs specific for the Rhesus D antigen was achieved by bio-panning on fresh whole rbc of phenotype R1R1 (CDe/CDe) i.e. the reference cells used for Rhesus D typing. This was necessary since the Rhesus D antigen, an integral membrane protein of 417 amino acids (Le Van Kim, C. et al, PNAS 89:10925, 1992), loses its immunogenicity during purification (Paradis, G. et al, J. Immunol. 137:240, 1986) and therefore a chemically purified D antigen

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cannot be bound to a solid phase for selection of immunoreactive Phabs as for other antigen specificities previously selected in this system (Vogel, M. et al., Eur.J. Immunol. 24:1200, 1994). Modelling studies have suggested that only very short connecting regions of the Rhesus D antigen extend outside 5 the cell membrane or protrude into the cytoplasm (Chérif-Zahar, B. et al, PNAS 87:6243, 1990). Thus the parts of the RhD antigen visible to antibodies are relatively restricted and may be under conformational constraint. This aspect of the Rhesus D antigen becomes even more important when considering selection of Phabs with reactivity against the partial D phenotypes which essentially lack certain defined epitopes of the D membrane protein (Mouro, I. et al, Blood. 83:1129, 1994).

Furthermore, since whole rbc do not only express the D antigen, a series of negative absorptions had to be performed on Rhesus D negative rbc in order to absorb out those Phabs reacting with the other antigenic proteins found on the rbc.

This panning procedure performed on Phabs coming from both LD1 and LD2 librairies resulted in the isolation of 7 different Fab producing clones from library LD1 and 9 different Fab producing clones from library LD2.

The nomenclature and the figures where the sequences are listed are given in table 1.

Table 1

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LIBRARY LD1 Clone No.	V _H - Sequence Figure	V _L - Sequence Figure	LIBRARY LD2 Clone No.	V _H - Sequence Figure	V _L - Sequence Figure
LD1-28	1a	1b	LD2-1	8a	8b
LD1-40	2a	2b	LD2-4	9a	9b
LD1-52	3 a	3b	LD2-5	10a	10b
LD1-84	4a	4b	LD2-10	11a	11b
LD1-98	5a	5b	LD2-11	12a	12b
LD1-110	6a	6b	LD2-14	13a	13b
LD1-117	7a	7b	LD2-17	14a	14b
			LD2-18	15a	15b
	·		LD2-20	16a	16b

The above Fab clones show exclusive reactivity against the Rhesus D antigen, 3 of 5 D^u rbc tested and agglutinating reactivity against the Partial D phenotypes as follows: Rh33, DIII, DIVa, DIVb, DVII.

However using the above mentioned R1R1 rbc for panning of the Phabs no clones were isolated which reacted against the Partial DVI phenotype. As the serum of the original hyperimmune donor tested at the time of construction of the recombinant library, was known to react against the DVI phenotype the recombinant library should also contain the anti-DVI specificity.

In order to select for the DVI reactivity the panning conditions were changed in that different cells were used. A special donor whose rbc had been typed and were known to express the Partial DVI phenotype was used as the source of cells for re-panning the LD1 and LD2 librairies. This second series of pannings was essentially performed in the same way as the first series except for the substitution of DVI rbc for R1R1 rbc and the addition of bromelase treatment to the DVI rbc. The DVI phenotype expresses the least number of Rhesus D epitopes and it is therefore difficult to make antibodies against it. It has been reported that only 15% of unselected polyclonal anti-D and 35% of selected anti-D made by Rhesus D negative subjects reacted with DVI+ cells (Mouro, I. et al, Blood, 83:1129, 1994). Bromelase treatment which removes N- acetylneuraminic acid (sialic acid) from the rbc membrane, was performed in order to render the Rhesus DVI epitopes more accessible during the panning with the pre-absorbed Phabs.

This second series of pannings on the LD1 and LD2 librairies resulted in 1 Fab producing clone reacting with DVI+ rbc.

The nomenclature is given below:

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LIBRARY LD1	V _H -Sequence figure	V _L -Sequence figure
Clone No: LD1-6-17	17a	17b

Thus a total of 17 different anti-Rhesus D Fab clones have been isolated. The DNA from these clones has been isolated and sequenced using Fluorescent Cycle Sequencing on an ABI 373A Sequencing System. The nucleotide and corresponding amino acid sequences of the said Fab clones form the basis of this invention.

The DNA sequences obtained and Fab fragments are useful for the preparation of complete antibodies having an activity against the Rhesus D antigen.

The examples which follow explain the invention in detail, without any restriction of the scope of the invention.

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Example 1 describes the construction of 2 combinatorial librairies; especially the aforementioned LD1 and LD2 libraries.

Example 2 describes a series of pannings using R1R1 rbc on the said LD1 and LD2 libraries in detail.

Example 3 describes a series of pannings using bromelase treated DVI+ rbc on the said LD1 and LD2 librairies.

Example 4 describes an indirect haemagglutination assay using a rabbit anti-phage antibody, as an equivalent Coombs reagent, to monitor the enrichment and specificity of Rhesus D specific Phabs after panning.

Example 5 describes the preparation and purification of Fab antibody fragments for application as diagnostic reagents.

Example 6 describes the preparation of complete anti-Rhesus D immunoglobulins using the sequences of the present invention.

Example 1

Construction of the recombinant LD1 and LD2 librairies

a) Source of the lymphocytes

A male adult who was a member of the volunteer pool of
hyperimmune Rhesus D donors was given an i.v. boost of 2 ml of packed rbc
from a known male donor of blood group O RhD⁺. The PBL were harvested at
+5 and +18 days after the boost and the mononuclear cells (MNC) isolated by
Ficoll gradient centrifugation (Lymphoprep, Pharmacia, Milwaukee, Wl). The
+5 day MNC were used directly for RNA preparation using a phenolchloroform guanidinium isothiocyanate procedure (Chomczynski, P. and
Sacchi, N., Anal. Biochem. 162:156, 1987). The +18 day MNC were first
cultured for 3 days in RPMI-1640 medium (Seromed, Basel) containing 10
U/ml of IL-2 (Sandoz Research Center, Vienna, Austria) and 10 μg/ml of
pokeweed mitogen (PWM; Sigma L9379, Buchs, Switzerland) before
extracting RNA.

Immunofluorescence analysis of donor lymphocytes +5 days after rbc i.v. boost

Cell Surface	% Positive	Cell Surface	% Positive
Antigen	cells	Antigen	cells .
CD20	15	CD8	12
CD38	20	CD25	7.6
CD20/38	15	CD57	12.5
CD3	47	CD14	6
CD4	34	HLA-DR	18

b) Construction of Library

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Two separate libraries were constructed called LD1 and LD2 (as detailed above) corresponding to the cells harvested at +5 days and +18 days (finally +21 days including the +3 days PWM stimulation) after the i.v. boost

respectively. Total RNA was then prepared from these cells using a phenolchloroform quanidinium isothiocyanate method. From this RNA, 10 μg were used to make cDNA using an oligo(dT) primer (400 ng) and reverse transcribed with M-MuLV reverse transcriptase according to the conditions specified by the supplier (Boehringer Mannheim Germany). PCR amplification was performed as described in Vogel, M. et al., E.J. of Immunol. 24:1200, 1994. Briefly, 100 μl PCR reaction contained Perkin-Elmer buffer with 10 mM MgCl₂, 5 μl cDNA, 150 ng of each appropriate 5' and 3' primer, all four dNTP at 200 µM each and 2 U/ml Tag Polymerase (Perkin Elmer, NJ). The PCR amplification of the heavy and light chains of the Fab molecule was performed separately with a set of primers from Stratacyte (details given below). For the heavy chain six upstream primers were used that hybridize to each of the six families of the V_H genes whereas one kappa and one lambda chain primer were used for the light chain. The downstream primers were designed to match the hinge region of the constant domains y1 and y3 for the heavy chain. For the light chain the downstream primers were matched to the 3' end of kappa and lambda constant domains. The heavy and light chain PCR products were pooled separately, gel purified and cut with Xho1/Spe1 and Sac1/ Xba1 restriction enzymes (Boehringer Mannheim), respectively. After digestion the PCR products were extracted once with phenol: chloroform: isoamylalcohol and purified by gel excision. The insertion of the Xho1/Spe1 digested Fd fragment and subsequent ligation of the Sac1/Xba1 digested light chain into the pComb3 vector, the transformation into XL1-Blue cells, and the production of phages were performed as described by (Barbas III, C.F. and Lerner, R.A., Companion Methods Enzymol. 2:119, 1991).

After transformation of the XL1-Blue E.coli cells samples were withdrawn and titrated on plates to determine the library size. These results indicated expression libraries of 7.5×10^6 and 7.7×10^6 cfu (colony forming units) for LD1 and LD2 respectively.

c) PCR Primers

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VHI 5'-CAC TCC CAG GTG CAG CTG CTC GAG TCT GG-3'
VHII 5'-GTG CTG TCC CAG GTC AAC TTA CTC GAG TCT GG-3'

VHIII 5'-GTC CAG GTG GAG GTG CAG CTG CTC GAG TCT GG-3'

VHIV 5'-GTC CTG TCC CAG GTG CAG CTG CTC GAG TCG GG-3'

VHV 5'-GTC TGT GCC GAG GTG CAG CTG CTC GAG TCT GG-3'

VHVI 5'-GTC CTG TCA CAG GTA CAG CTG CTC GAG TCA GG-3'

5 CHI(gI) 5'-AGC ATC ACT AGT ACA AGA TTT GGG CTC-3'

- VL(k) 5'-GT GCG AGA TGT GAG CTC GTG ATG ACC CAG TCT CCA-3'
- CL(k) 5'-T CCT TCT AGA TTA CTA ACA CTC TCC CCT GTT GAA GCT CTT TGT GAC GGG CGA ACT C-3'
- VL(I) 5'C TGC ACA GGG TCC TGG GCC GAG CTC GTG GTG ACT CA-3'
- O CL(I) 5'G CAT TCT AGA CTA TTA TGA ACA TTC TGT AGG GGC-3'

d) Vectors and bacterial strains

The pComb3 vector used for cloning of the Fd and the light chain was obtained from the Scripps Research Institute La Jolla, CA; (Barbas III, C.F. and Lerner, R.A., Companion Methods Enzymol. 2:119, 1991). The *Escherichia coli* strain XL1-Blue used for transformation of the pComb3 vector and the VCSM13 helper phage were purchased from Stratacyte (La Jolla, CA).

Example 2

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Selection of Rhesus D Phabs from LD1 and LD2 librairies on R1R1 rbc

a) Absorption and Bio-Panning

A series of three negative absorptions on rbc group O Rh negative were performed for each panning round before positive selection on rbc group O Rh positive (R1R1). Fresh rbc were collected in ACD (acid citrate dextrose) anticoagulant and washed 3 times in 0.9% NaCl. The rbc were counted in Hayems solution and adjusted to 40x10⁶/ml. Absorption : 1 ml of phage preparation in PBS/3%BSA was added to rbc group O Rh negative pellet (16x10⁶ rbc) in 12 ml tubes (Greiner 187261, Reinach, Switzerland) and incubated at RT for 30 min. with careful shaking. All tubes were pre-blocked in PBS/3% BSA for a minimum of 1hr at RT. The rbc were pelleted by centrifuging for 5 min. 300 x g at 4°C. The resulting phage supernatant was

carefully harvested and the process repeated twice more. After the final absorption the phage supernatant was added to the rbc group O Rh positive pellet (16×10^6 rbc) and again incubated at RT for 30 min. with gentle shaking. Then the rbc were washed at least 5 times in 10 ml ice cold PBS, centrifuged 5 min. $300\times g$ at 4°C, followed by elution with $200\ \mu l$ of 76 mM citric acid pH 2.8 for 6 min. at R.T. and neutralisation with $200\ \mu l$ 1M Tris. The rbc were centrifuged $300\times g$, 5 min. at 4°C and the resulting supernatant containing the eluted phages was carefully removed and stored with carrier protein (0.3% BSA) at 4°C ready for re-amplification.

Selection of Rhesus D+ Phabs from the LD1 and LD2 librairies on R1R1 rbc

Panning	No. of eluted Rhesus D	specific phages	
Round No.a)	Library D1 cfu	Library D2 cfu	
1	8x10 ⁶	4.6x10 ⁷	
2	6x10 ⁷	1.4x10 ⁷	
3	1x10 ⁸	7.9x10 ⁷	
4	3x10 ⁸	1.3x10 ⁸	
5	3x10 ⁸	1x10 ⁸	
6	nd	2.8x10 ⁸	

a) For each round 10¹² Phabs were incubated in tubes with rbc Group O Rhesus negative (absorption phase) followed by elution from rbc Group O Rhesus positive (R1R1)

nd = not done

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cfu = colony forming units

Example 3

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Selection of Rhesus D Phabs from LD1 and LD2 librairies on DVI+ rbc

a) Absorption on rbc group O Rh negative

A series of three negative absorptions on rbc group O Rh negative were performed for each panning round before positive selection on rbc group O Rh DVI positive. Fresh rbc were collected in ACD anticoagulant and washed 3 times in 0.9% NaCl. The rbc were counted in Hayems solution and adjusted to 40×10^6 /ml. Absorption: 1 ml of phage preparation in PBS/3%BSA was added to rbc group O Rh negative pellet (16×10^6) in 12 ml tubes (Greiner 187261, Reinach, Switzerland) and incubated at RT for 30 min. with careful shaking. All tubes were pre-blocked in PBS/3% BSA for a minimum of 1hr at RT. The rbc were pelleted by centrifuging for 5 min. 300 x g at 4°C. The resulting phage supernatant was carefully harvested and the process repeated twice more. Treatment of the rbc group O Rhesus DVI+ with the enzyme bromelase was performed at this stage in order to enhance accessibility of the antigens.

b) Treatment of rbc Rhesus DVI+ with bromelase

Bromelase 30 (Baxter, Düdingen, Switzerland) was used to treat rbc Rhesus DVI+ in the same proportions as used in a routine haemagglutination assay, i.e. 10 μ l bromelase per 2x10⁶ rbc. Thus 80 μ l of bromelase was added to 16x10⁶ DVI+ rbc and incubated at 37°C for 30 min. followed by washing 3 times in 0.9% NaCl, re-counting in Hayems solution and adjusting to 40x10⁶/ml in PBS/3% BSA ready for Phab panning.

c) Bio-Panning on bromelase treated Rhesus DVI+ rbc

After the final absorption the phage supernatant was added to the enzyme treated rbc group O Rh DVI+ pellet $(16x10^6)$ and again incubated at RT for 30 min. with gentle shaking. Then the rbc were washed at least 5 times in 10 ml ice cold PBS, centrifuged 5 min. 300 x g at 4°C, followed by elution with 200 μ l of 76 mM citric acid pH 2.8 for 6 min. at R.T. and neutralisation with 200 μ l 1M Tris. The rbc were centrifuged 300 x g, 5 min. at 4°C and the

resulting supernatant containing the eluted phages was carefully removed and stored with carrier protein (0.3% BSA) at 4°C ready for re-amplification.

Selection of Rhesus D Phabs from LD1 and LD2 librairies on Rhesus DVI+ red blood cells

Panning	No. of eluted Rhesus D	specific phages		
Round No. ^{a)}	Library LD1 cfu	Library LD2 cfu		
1	5x10 ⁷	4.6x10 ⁷		
2	1.8x10 ⁷	1.4×10 ⁷		
3	4x10 ⁸	7.9x10 ⁷		
4	6.8x10 ⁸	1.3×10 ⁸		
5	5.8x10 ⁸	1×10 ⁸		

a) For each round 10¹² Phabs were incubated in tubes with rbc Group O Rhesus negative (absorption phase) followed by elution from rbc Group O Rhesus DVI+

Example 4

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Monitoring of the panning rounds and determination of the specificity of the enriched Phabs using a rabbit anti-phage antibody

Indirect haemagglutination assay

Freshly collected rbc of different ABO and Rhesus blood groups were washed 3 times in 0.9% NaCl and adjusted to a 3-5% solution (45- $50x10^7$ /ml) in either 0.9% NaCl or PBS/3% BSA. For each test condition 50 μ l rbc and 100 μ l test (precipitated and amplified phage or control antibodies) were incubated together in glass blood grouping tubes (Baxter, Düdingen, Switzerland) for 30 min. at 37°C. The rbc were washed 3 times in 0.9% NaCl and then incubated with 2 drops of Coombs reagent (Baxter, Düdingen, Switzerland) for positive controls or with 100 μ l of 1/1000 diluted rabbit antiphage antibodies (made by immunising rabbits with phage VCSM13

preparation, followed by purification on an Affi-Gel Blue column and absorption on E. coli to remove E. coli-specific antibodies). The tubes were incubated for 20 min at 37°C, centrifuged 1min at 125xg and rbc examined for agglutination by careful shaking and using a magnifier viewer.

When purified Fab were tested for agglutination an affinity purified anti-Fab antibody (The Binding Site, Birmingham, U.K.) was used instead of the rabbit anti-phage antibody.

Monitoring of Phabs from LD1 and LD2 librairies by indirect haemagglutination after Panning on R1R1 rbc

Phab sample Panning Round	Library LD1 Library LD2 tested on rbc O Rh D+ (a)				
No. 4					
undiluted	+	+			
1/4	+	+/-			
1/20	-	-			
No.5					
undiluted	++	+			
1/4	++	+			
1/20	-	-			
No. 6					
undiluted	nd	+++			
1/4	nd	++			
1/20	nd	nd ·			
Helper phage (b)					
undiluted, 1/4, 1/20	-	-			

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- a) Indirect haemagglutination was performed in glass tubes using 50 μ l rbc (40x10 7 /ml) and 100 μ l Phabs starting at 4x10 11 /ml. After 30 min. at 37°C the rbc were washed 3 times and further incubated for 20 min. at 37°C with a 1/1000 dilution of rabbit anti-phage antibody.
- b) The M13 helper phage was used as a negative control and showed no non-specific agglutination due to the phage particle alone.

Agglutination was scored by visual assessment from +++ (strong agglutination) descending to - (no agglutination). nd = not done

Reaction Pattern of Fab Rhesus D Clones, resulting from R1R1 rbc panning, against Partial D Variants

		F	Partial D V	ariants		
(a) Reaction Pattern of Clones from Library LD2	Rh33	DIII	DIVa	DIVb	DVI	DVII
Pattern 1 n=2 (b)	+++	nd	+++	+++	_	+++
Pattern 2 n=1	-	+++	-	+	-	+++
Pattern 3 n=1	-	+++	+++	+	-	+++
Pattern 4 n=2	-	nd	+++	+++	-	+++
Pattern 5 n=1	+++	+++	+++	+++	-	+++
Pattern 6 n=1	-	+++	-	-	-	+++
Pattern 7 n=1	-	+++	+++	-	-	+++

- a) soluble Fab preparations were made of each clone (as detailed in example 5) followed by indirect haemagglutination on the above panel of D variants
- b) n represents number of clones in each reaction pattern

Pattern 1:

clones: LD2-1.14

Pattern 2:

clone:

LD2-4 clone: LD2-17

Pattern 3: 10 Pattern 4:

clones: LD2-5,10

Pattern 5:

clone:

LD2-18

Pattern 6:

clone:

LD2-11

Pattern 7:

clone:

LD2-20

nd = not done

Reaction Pattern of Rhesus D Clones against Partial DVI Variant

Only the LD1-6-17 clone resulting from panning against DVI+rbc reacted with DVI+ and R1R1 rbc in the indirect haemagglutination assay using rabbit anti phage antibody as the Coombs equivalent reagent and in the presence of Enlisst (Baxter).

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Example 5

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Preparation and purification of Fab antibody fragments for application as diagnostic reagents

After the bio-panning procedures detailed in Examples 2 and 3 a phage population which showed specific agglutination on Rhesus D+ rbc was selected and used to prepare phagemid DNA. More precisely the Phabs selected on R1R1 rbc were used after the 5th and 6th rounds of bio-panning for LD1 and LD2 libraries respectively and after the 5th bio-panning on DVI+ rbc for isolation of the LD1-6-17 clone. In order to produce soluble Fab, the sequence gIII coding for the pIII tail protein of the phage particle must be deleted.

Phagemid DNA was prepared using a Nucleotrap kit (Machery-Nagel) and the gIII sequence was removed by digesting the so isolated phagemid DNA with Nhe1/Spe1 as described (Burton, D.R., et al., PNAS, 1989). After transformation into XL1-Blue individual clones were selected (nomenclature given in table 1) and grown in LB (Luria Broth) containing 50 μg/ml carbenicillin at 37°C to an OD of 0.6 at 600 nm. Cultures were induced with 2 mM isopropyl β-D-thiogalactopyranoside (IPTG) (Biofinex, Praroman, Switzerland) and grown overnight at 37°C. The whole culture was spun at 10,000xg for 30 min. at 4°C to pellet the bacteria. The bacterial pellet was treated with a lysozyme/DNase solution to liberate the Fab fragments inside the cells. As some Fab were released into the culture supernatant this was also harvested separately. These Fab preparations were then pooled and precipitated with 60% ammonium sulphate (Merck, Darmstadt, Germany) to concentrate the Fab followed by extensive dialysis in phosphate buffered saline (PBS) and ultracentrifugation at 200,000xg to pellet any insoluble complexes. The Fab preparations were then purified on a ceramic hydroxyapatite column (HTP Econo cartridge, BioRad, Glattbrugg, Switzerland) using a gradient elution of PBS (Buffer A) and PBS + 0.5M NaCl 30 (Buffer B). The linear gradient was programmed to increase from 0-100% Buffer B in 40 min. The Fab was eluted as a single peak between 40-60% Buffer B. The positive fractions as identified by immunodot assay using an

anti-Fab peroxidase conjugate (The Binding Site, Birmingham, U.K.) were pooled, concentrated using polyethylene glycol and extensively dialysed against PBS. The positive fractions from the hydroxyapatite column for each clone were used in a classical indirect haemagglutination assay in glass tubes using either the standard Coombs reagent (Baxter Diagnostics AG Dade, anti-human serum) or an anti-Fab (The Binding Site, Birmingham, U.K.) as the cross linking reagent. These Fab of defined specificity on the Partial D variants as shown on page 18 can be used to type rbc of unknown Partial D phenotype.

10 Example 6

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Construction of complete immunoglobulin genes

The LD2-14 heavy chain V gene (VH gene) was amplified from the anti-Rhesus D-Fab-encoding plasmid LD2-14 with the polymerase chain reaction (PCR) using gene-specific primers. The 5'-primer had the sequence 5'-AGGTGTCGACGCACAGGTGAAACTGCTCGAG-3' whereas the 3'-primer was of the sequence: 5'-GAGGAGACGGTGACCGTGGT-3'. The PCR reaction was performed with Tag DNA Polymerase and the buffer solution from Boehringer Mannheim (Mannheim, Germany) at the conditions recommended by the manufacturer including 60 pmol of each primer and the four deoxynucleotides at a concentration of 0.25 μM each. The reaction was run for 35 cycles with the following temperature steps: 60 s at 94°C (extended by 5 min. during the first cycle), 75 s at 55°C and 90 s at 72°C (extended by 5 min. during the last cycle). The PCR product was purified with the QIAquick kit from Qiagen, Basel, Switzerland, digested with the restriction enzymes Sal I and BstE II, extracted with phenol, purified by preparative agarose gel electrophoresis, concentrated by precipitation with ethanol and solubilized in a minimal volume of LTE buffer (10 mM Tris-HCl pH 8.0, 0.1 mM EDTA).

Vector # 150 (Sandoz Pharma, Basel) which contained an irrelevant intact human genomic immunoglobulin V_H gene was cut with *Sal* I and *Bst*E II, treated with calf intestinal phosphatase, extracted twice with

phenol, once with chloroform/isoamylalcohol (24:1), concentrated by precipitation with ethanol and solubilized in a minimal volume of LTE. 100 ng of this vector and 15 ng of the digested and purified PCR product were ligated according to the recommendations of the manufacturer in a total volume of 20 μl (Ligase and buffer concentrate from Boehringer Mannheim (Mannheim)). The ligase was inactivated for 10 min. at 65°C, the reaction mix diluted to 100 μl with H_20 and 3 μl of the diluted solution were electroporated with a GenePulser (BioRad, Gaithersburg) into 40 μl of competent E. coli JA221 according to the recommendations of the manufacturer (0.1 cm cuvettes, 1.8 kV, 200 Ω , 25 μFD), diluted with SOC medium, incubated at 37°C without shaking for 1 h and plated on LB plates containing ampicillin (50 $\mu g/ml$). Plasmid-minipreps of the resulting colonies were checked with restriction digests for the presence of the appropriate insert.

With this procedure, the irrelevant resident V_H gene in vector # 150 was replaced by the amplified anti-Rhesus D V_H sequence and yielded plasmid # 150-LD2-14. The structure of the resulting genomic immunoglobulin V_H gene construct was confirmed by sequencing, cut out by digestion with *EcoR* I and *BamH* I and gel purified as described above for the PCR product. Expression vector # 10 (Sandoz Pharma, Basel) containing the human genomic immunoglobulin C₂1 gene segment was also digested with *EcoR* I and *BamH* I, treated with calf intestinal phosphatase, extracted with phenol, concentrated by precipitation with ethanol as described above, ligated with the *EcoR* I/BamH I-V_H gene segments previously obtained from plasmids # 150-LD2-14 and transfected into E. coli JA221 as described above. This second cloning step yielded a complete anti-Rhesus D heavy chain immunoglobulin gene in plasmid # 10-LD2-14 (Figures 19 and 20).

The LD2-14 light chain V gene (V_L gene) was amplified from the same anti-Rhesus D-Fab plasmid LD2-14 by PCR using gene-specific primers. The 5'-primer had the sequence: 5'-GGTACGCGTTGTGAGCTCGTGATGACCCAG-3' whereas the 3'-primer was of the sequence 5'-TTTGATCTCAAGCTTGGTCCCAGGGCC-3'.

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PCR reaction, product purification and subsequent cloning steps were performed as described for the V_H gene, except that the appropriate light chain vectors were used. Briefly, the V₁ PCR product was digested with the restriction enzymes Mlul and Hind III, extracted with phenol, purified from an agarose gel, ligated into vector # 151 (Sandoz Pharma, Basel) and propagated in E. coli JA221. This vector had been cut with Mlul and Hind III, treated with calf intestinal phosphatase, extracted with phenol and concentrated by precipitation with ethanol. Plasmid # 151 contained an irrelevant intact human genomic immunoglobulin V_L gene. With this procedure, the irrelevant resident V₁ gene was replaced by the amplified anti-Rhesus D sequence and yielded plasmids # 151-LD2-14. The structure of the resulting genomic construct was confirmed by sequencing, cut out by digestion with EcoR I and Xba I and gel purified as described above. Subsequently, expression vector # 98 (Sandoz Pharma, Basel, Switzerland) containing the human genomic immunoglobulin Cκ gene segment was digested with EcoR I and Xba I, treated with calf intestinal phosphatase, extracted with phenol, concentrated by precipitation with ethanol, ligated with the EcoR I/Xba I-V_L gene segment obtained from plasmid # 151-LD2-14 and transfected into E. coli JA221. This procedure yielded a complete anti-Rhesus D light chain immunoglobulin gene in plasmid # 98-LD2-14.

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Restriction digests confirmed the structure of the expression constructs for LD2-14 heavy and light chain. The mouse myeloma cell line SP2/0-Ag 14 (ATCC CRL 1581) was cotransfected by electroporation with the plasmids # 10-LD2-14 and # 98-LD2-14. The electroporation was performed as follows: Exponentially growing cells were washed twice and suspended in phosphate buffered sucrose (272 mM sucrose, 1 mM MgCl₂, 7 mM NaH₂PO₄, pH 7.4) at a density of 2 x 10⁷ cells/ml. 0.8 ml of cells were added to a 0.4 cm cuvette, mixed with 15 μg of linearized plasmids # 10-LD2-14 and # 98-LD2-14, held on ice for 15 min., electroporated with 290 Volts, 200 Ω , 25 μFD , put back on ice for 15 min., transferred to a T75 cell culture flask with 20 ml of cold RPMI 1640 medium (10% heat inactivated fetal bovine serum, 50 μM beta-mercaptoethanol), left for 2 h at room temperature and then incubated for 60 h at 37°C. After this period, the cells were transferred to 50 ml of medium containing 1 mg/ml G418 for selection. Two weeks later, all non-

transformed cells had died and the supernatants tested positive for the presence of human IgG/k by an enzyme linked immuno-sorbent assay (ELISA). The cultures were cloned by limiting dilution in microtiter plates and the supernatants quantitated by ELISA. The best producer clone was used for production cultures. From 2 - 3 liters of culture supernatant the antibodies were purified by affinity chromatography on a Protein G Sepharose (Pharmacia, Uppsala) according to the recommendations of the manufacturer. Briefly, the supernatant was diluted with 1 volume of 10 mM phosphate buffer pH 7.0, 0.02% NaN₃ and pumped over a column with 2 ml of Protein G Sepharose. The column was washed with 30 ml of 10 mM phosphate buffer pH 7.0, 0.02% NaN₃ and the bound antibodies were eluted with 0.1 M Glycine-HCl pH 2.0, 0.02% NaN₃ and immediately neutralised with solid NaHCO₃. These antibodies (rD2-14) were positive for agglutination of Rhesus D positive and negative for Rhesus D negative red blood cells.

The same procedure was used to produce complete recombinant antibodies from the other clones from LD1 and LD2. Because of sequence diversity, for some clones different PCR primers were synthesized. For the heavy chain this was LD1-84, where the sequence of the 5'-primer was 5'-AGGTGTCGACGCACAGGTAAACTGCTCGAG-3'.) For the light chain the 5'-primer was the same as for LD2-14 noted above (except for LD2-1, -10, where the sequence was 5'-GGTACGCGTTGTGA-GCTCGTGGTGACTCAG-3') whereas the 3'-primer was of the sequence 5'-TTTGATTTCAAGC-TTGGTCCCTTGGCC-3', except for some clones where the following 3'-primers were used:

25 LD1-52, -98: 5'-TTTGATCTCAAGCTTGGTCCCAGGGCC-3',

LD1-40: 5'-TTTGATCTCAAGCTTTGTCCCTTGGCC-3',

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LD1-84: 5'-TTTGATGTCAAGCTTGGTCCCCCGGCC-3',

LD2-11: 5'-TTTGATCTGAAGCTTGGTCCCCTGCCC-3'.

LD2-1, -10: 5'-TAGGACGGTAAGCTTGGTCCCTCCGCC-3').

The cloning steps for the production of complete antibody heavy and light chain genes were the same as those described above for LD2-14. Expression in SP2/0-Ag14 cells yielded antibodies with the same anti-Rhesus D specificities as the Fab expressed in E. coli.

Claims

1. Polypeptides capable of forming antigen binding structures with specificity for Rhesus D antigens which include Rhesus D-specific CDR 1, CDR 2 and CDR 3 regions of the amino acid sequences V_{H} and V_{L} with the identification numbers according to the figures given in the table below:

	V _H				VL			
Identi-		CDR 1	CDR 2	CDR 3	_	CDR 1	CDR 2	CDR 3
fication	Figure	base pair	base pair	base pair	Figure	base pair	base pair	base pair
No.		-No.	No.	No.		No.	No.	No.
LD1-28	Fig. 1a	91-104	148-198	295-342	Fig. 1b	64-96	142-162	259-288
LD1-40	Fig. 2a	91-105	148-198	295-342	Fig. 2b	64-96	142-162	259-288
LD1-52	Fig. 3a	91-105	148-198	295-342	Fig. 3b	64-96	142-162	259-288
LD1-84	Fig. 4a	91-105	148-198	295-342	Fig. 4b	64-96	142-162	259-285
LD1-98	Fig. 5a	91-105	148-198	295-342	Fig. 5b	64-96	142-162	259-288
LD1-110	Fig. 6a	91-105	148-198	295-342	Fig. 6b	64-96	142-162	259-285
LD1-117	Fig. 7a	91-105	148-198	295-345	Fig. 7b	64-96	142-162	259-288
LD2-1	Fig. 8a	91-105	148-198	295-342	Fig. 8b	61-99	145-165	262-294
LD2-4	Fig. 9a	91-105	148-198	295-342	Fig. 9b	64-96	142-162	259-282
LD2-5	Fig.10a	91-105	148-198	295-342	Fig. 10b	64-96	142-162	259-288
LD2-10	Fig. 11a	91-105	148-198	298-345	Fig. 11b	61-102	148-168	265-294
LD2-11	Fig. 12a	91-105	148-198	295-342	Fig. 12b	64-96	142-162	259-285
LD2-14	Fig. 13a	91-105	148-198	295-342	Fig. 13b	64-96	142-162	259-285
LD2-17	Fig. 14a	91-105	148-198	295-342	Fig. 14b	64-96	142-162	259-285
LD2-18	Fig. 15a	91-105	148-198	295-342	Fig. 15b	64-96	142-162	259-288
LD2-20	Fig. 16a	91-105	148-198	295-342	Fig. 16b	64-96	142-162	259-285
LD1-6-17	Fig. 17a	91-105	148-198	295-351	Fig. 17b	64-96	142-162	259-285

2. Polypeptides according to claim 1 which include regions with the amino acid sequences V_H and V_L and have identification numbers according to the figures given in the table of claim 1.

- 3. Polypeptides according to claim 1 or 2 characterised as antigen binding Fab fragments.
- 4. Polypeptides according to claim 1 or 2 comprising immunoglobulin heavy and light chains capable of forming complete anti-Rhesus D antibodies.
- 5. DNA sequences coding for polypeptides capable of forming antigen binding structures with specificity for Rhesus D antigens which include regions with the Rhesus D-specific CDR 1, CDR 2 and CDR 3 segments of the DNA sequences V_H and V_L with the identification numbers according to the figures given in the table below, and functional equivalents thereof:

	V _H			V _L				
ldenti-		CDR 1	CDR 2	CDR 3		CDR 1	CDR 2	CDR 3
fication	Figure	base pair	base pair	base pair	Figure	base pair	base pair	base pair
No.		No.	No.	No.		No.	No.	No.
LD1-28	Fig. 1a	91-104	148-198	295-342	Fig. 1b	64-96	142-162	259-288
LD1-40	Fig. 2a	91-105	148-198	295-342	Fig. 2b	64-96	142-162	259-288
LD1-52	Fig. 3a	91-105	148-198	295-342	Fig. 3b	64-96	142-162	259-288
LD1-84	Fig. 4a	91-105	148-198	295-342	Fig. 4b	64-96	142-162	259-285
LD1-98	Fig. 5a	91-105	148-198	295-342	Fig. 5b	64-96	142-162	259-288
LD1-110	Fig. 6a	91-105	148-198	295-342	Fig. 6b	64-96	142-162	259-285
LD1-117	Fig. 7a	91-105	148-198	295-345	Fig. 7b	64-96	142-162	259-288
LD2-1	Fig. 8a	91-105	148-198	295-342	Fig. 8b	61-99	145-165	262-294
LD2-4	Fig. 9a	91-105	148-198	295-342	Fig. 9b	64-96	142-162	259-282
LD2-5	Fig.10a	91-105	148-198	295-342	Fig. 10b	64-96	142-162	259-288
LD2-10	Fig. 11a	91-105	148-198	298-345	Fig. 11b	61-102	148-168	265-294
LD2-11	Fig. 12a	91-105	148-198	295-342	Fig. 12b	64-96	142-162	259-285
LD2-14	Fig. 13a	91-105	148-198	295-342	Fig. 13b	64-96	142-162	259-285
LD2-17	Fig. 14a	91-105	148-198	295-342	Fig. 14b	64-96	142-162	259-285
LD2-18	Fig. 15a	91-105	148-198	295-342	Fig. 15b	64-96	142-162	259-288
LD2-20	Fig. 16a	91-105	148-198	295-342	Fig. 16b	64-96	142-162	259-285
LD1-6-17	Fig. 17a	91-105	148-198	295-351	Fig. 17b	64-96	142-162	259-285

- 6. DNA sequences according to claim 5 which include regions with the DNA sequences V_H and V_L with the identification numbers according to the figures given in claim 5.
- 7. DNA sequences according to claim 5 or 6 coding for polypeptides capable of forming antigen binding Fab fragments.
 - 8. DNA sequences according to claim 5 or 6 coding for polypeptides capable of forming complete anti-Rhesus D antibodies.
- 9. A process for preparing recombinant polypeptides capable of forming antigen binding structures, e.g. Fab fragments, with specificity for Rhesus D antigens which process comprises the following steps in sequential order:
 - a) boosting of an individual capable of forming anti-Rhesus D antibodies with Rhesus D positive red blood cells,
 - b) isolating mononuclear cells from the individual,
- c) isolating total RNA from the mononuclear cells,
 - d) preparing a cDNA by using an oligo(dT)primer and reverse transcribing of the mRNA with M-MuLV reverse transcriptase and amplifying the cDNA repertoire by a polymerase chain reaction using immunoglobulin gene family specific primers,
 - e) creating a phage display library by inserting the DNA coding for the heavy and light chain of the Fab polypeptide into a phagemid vector; the DNA for the heavy chain is inserted in frame to the gene coding for the phage protein pIII which allows the expression of a Fab pIII fusion protein on the surface of the phage,
- f) transforming bacterial cells with the obtained recombinant plasmids, cultivating of the transformed bacterial cells and co-expression of the heavy and the light chain of a Fab on filamentous phage particles,
 - g) amplifying the Fab-carrying phage in bacteria,

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 h) selecting individual phage clones by several rounds of panning on Rhesus positive red blood cells.

- i) isolating the plasmid DNA from the selected clones and cutting out the gIII gene,
- j) transforming bacterial cells with the obtained plasmid, cultivating of the transformed bacterial cells expressing the Fab, and isolating the Fab fragments.

10. Anti-Rhesus D antibodies having heavy and light chain variable regions comprising the Rhesus D-specific CDR 1, CDR 2 and CDR 3 sequences of the amino acid sequences V_H and V_L given the identification numbers as indicated in the table below:

	V _H				V _L			
ldenti-		CDR 1	CDR 2	CDR 3		CDR 1	CDR 2	CDR 3
fication	Figure	base pair	base pair	base pair	Figure	base pair	base pair	base pair
No.		No.	No.	No.		No.	No.	No.
LD1-28	Fig. 1a	91-104	148-198	295-342	Fig. 1b	64-96	142-162	259-288
LD1-40	Fig. 2a	91-105	148-198	295-342	Fig. 2b	64-96	142-162	259-288
LD1-52	Fig. 3a	91-105	148-198	295-342	Fig. 3b	64-96	142-162	259-288
LD1-84	Fig. 4a	91-105	148-198	295-342	Fig. 4b	64-96	142-162	259-285
LD1-98	Fig. 5a	91-105	148-198	295-342	Fig. 5b	64-96	142-162	259-288
LD1-110	Fig. 6a	91-105	148-198	295-342	Fig. 6b	64-96	142-162	259-285
LD1-117	Fig. 7a	91-105	148-198	295-345	Fig. 7b	64-96	142-162	259-288
LD2-1	Fig. 8a	91-105	148-198	295-342	Fig. 8b	61-99	145-165	262-294
LD2-4	Fig. 9a	91-105	148-198	295-342	Fig. 9b	64-96	142-162	259-282
LD2-5	Fig.10a	91-105	148-198	295-342	Fig. 10b	64-96	142-162	259-288
LD2-10	Fig. 11a	91-105	148-198	298-345	Fig. 11b	61-102	148-168	265-294
LD2-11	Fig. 12a	91-105	148-198	295-342	Fig. 12b	64-96	142-162	259-285
LD2-14	Fig. 13a	91-105	148-198	295-342	Fig. 13b	64-96	142-162	259-285
LD2-17	Fig. 14a	91-105	148-198	295-342	Fig. 14b	64-96	142-162	259-285
LD2-18	Fig. 15a	91-105	148-198	295-342	Fig. 15b	64-96	142-162	259-288
LD2-20	Fig. 16a	91-105	148-198	295-342	Fig. 16b	64-96	142-162	259-285
LD1-6-17	Fig. 17a	91-105	148-198	295-351	Fig. 17b	64-96	142-162	259-285

- 11. Anti-Rhesus D antibodies according to claim 10 with the amino acid sequences V_H and V_L and the identification numbers according to the figures, as indicated in the table of claim 10.
- 12. Anti-Rhesus D antibodies according to claim 10 or 11 wherein the immunoglobulin constant regions are of at least one of the defined isotypes IgG1, IgG2, IgG3 or IgG4.
 - 13. A process for preparing complete anti-Rhesus D antibodies according to one of the claims 10 to 12, comprising in sequential order the steps of
- a) amplifying separately the members of a pair of a heavy chain V gene segment and a light chain V gene segment containing Rhesus D-specific CDR 1, CDR 2 and CDR 3 regions as depicted in Figs. 1a 17a and 1b 17b, respectively, from an anti-Rhesus D-Fab-encoding plasmid by carrying out a polymerase chain reaction with specific primers,

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- b) preparing separately the genes of a complete anti-Rhesus D immunoglobulin heavy chain and a complete anti-Rhesus D immunoglobulin light chain in suitable plasmids containing the immunoglobulin constant region gene segments coding for either one of the human $\gamma 1$, $\gamma 2$, $\gamma 3$ and $\gamma 4$ heavy chains and for the human κ or λ light chain and transforming the obtained plasmids separately in suitable E. coli bacteria, and
- c) cotransfecting the obtained plasmids into a suitable mouse myeloma cell line, cultivating of the cells, separating the non-transformed cells, cloning of the cultures, selecting the best producing clone, using it as a production culture and isolating the complete antibodies from the supernatant of the cell culture.
- 14. A pharmaceutical composition comprising at least one polypeptide according to the definition of claim 1 or 2 or at least one anti-Rhesus D antibody according to one of the claims 10 to 12 for the prophylaxis

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of haemolytic disease of the newborn, for the treatment of idiopathic thrombocytopenic purpura and mistransfusions of Rhesus incompatible blood.

15. A diagnostic composition for Rhesus D typing comprising Fab fragments according to claim 3 or anti-Rhesus D antibodies according to one of the claims 10 to 12.

~~10 DOC

Abstract

Polypeptides capable of forming antigen binding structures specific for Rhesus D antigens include the sequences indicated in the figs. 1a to 17 b. They are prepared in a process comprising the following steps in sequential order:

- boosting of an individual capable of forming anti-Rhesus D antibodies with Rhesus D positive red blood cells,
- isolating mononuclear cells from the individual,
- isolating total RNA from the mononuclear cells,
- preparing a cDNA by using an oligo(dT)primer and reverse transcribing of the mRNA with M-MuLV reverse transcriptase and amplifying the cDNA repertoire by a polymerase chain reaction using immunoglobulin gene family specific primers,
- creating a phage display library by inserting the DNA coding for the heavy and light chain of the Fab polypeptide into a phagemid vector; the DNA for the heavy chain is inserted in frame into the gene coding for the phage protein pIII which allows the expression of a Fab pIII fusion protein on the surface of the phage,
- transforming bacterial cells with the obtained recombinant plasmids, cultivation of the transformed bacterial cells and coexpression of the heavy and the light chain of a Fab on filamentous phage particles,
- amplifying the Fab-carrying phage in bacteria,
- selecting individual phage clones by several rounds of panning on Rhesus positive red blood cells,
- isolating the plasmid DNA from the selected clones and cutting out the glll gene,
- transforming bacterial cells with the obtained plasmid, cultivating of the transformed bacterial cells expressing the Fab, and isolating the Fab fragments.

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The obtained polypeptides, being Fab fragments, can be used directly as an active ingredient in pharmaceutical and diagnostic compositions. The Fab and their DNA sequences can also be used for the preparation of complete recombinant Anti-Rhesus D antibodies.

- (1) GENERAL INFORMATION
- (i) APPLICANT:
- (A) NAME: Rotkreuzstiftung Zentrallaboratorium Blutspendedienst SRK
- (B) STREET: Wankdorfstrasse 10
- (C) CITY: Bern 22
- (D) STATE OR PROVINCE:
- (E) COUNTRY: Switzerland
- (F) POSTAL CODE: CH-3000
- (ii) TITLE OF INVENTION: Polypeptides capable of forming antigen binding structures with specificity for the Rhesus D antigens, the DNA encoding them and the process for their preparation and use
- (iii) NUMBER OF SEQUENCES: 34
- (iv) COMPUTER-READABLE FORM:
- (A) MEDIUM TYPE: 3.5" Floppy disk, 1.44 MB
- (B) COMPUTER: IBM compatible PC
- (C) OPERATING SYSTEM: IBM-DOS 6.3/Windows 3.1
- (D) SOFTWARE: MS Word for Windows 6.0 /saved as MS-DOS text
- (v) CURRENT APPLICATION DATA: n.a.
- (A) APPLICATION NUMBER: n.a.

- (2) INFORMATION FOR SEQ ID NO: 1 (LD1-28-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD1
- (B) CLONE: LD1-28
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

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(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 1 (LD1-28-VH)

		GAG Glu						48
		TGT Cys						96
		CGC Arg						144
		GAT Asp						192
		ATC Ile 70						240
		CTG Leu						288
		GTT Val						336
		GGG Gly						375

- (2) INFORMATION FOR SEQ ID NO:2 (LD1-28-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-28
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 2 (LD1-28-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AAC ATT ATC CGC TAT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Asn Ile Ile Arg Tyr Leu Asn 20 25 TGG TAT CAG CAG AAG CCA GGG AAA GCC CCT AGG CTC CTG ATC TAT GGT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Arg Leu Leu Ile Tyr Gly 40 GCG TCC ACT TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA Ala Ser Thr Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 50 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGT AGT CTG CAA CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp TTT GCA ACT TAC TAC TGT CAA CAG AGT TAC CGT ACC CCT CCA TTC ACT Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Arg Thr Pro Pro Phe Thr 85 TTC GGC CCT GGG ACC AAA GTG GAG ATC AAA 318 Phe Gly Pro Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 3 (LD1-40-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-40
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q 32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 3 (LD1-40-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCT GGG AGG Gln Val Lys Leu Leu Glu Ser Gly Gly Val Val Gln Pro Gly Arg 10 TCC CTG AGA CTC TCC TGT ATA GCG TCT GGA TTC ACC CTC AGG AAT TAT Ser Leu Arg Leu Ser Cys Ile Ala Ser Gly Phe Thr Leu Arg Asn Tyr 20 GCC ATG CAC TGG GTC CGC CAG GCT CCA GGC AAG GGG CTG GAG TGG GTG 144 Ala Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val GCA GGT ATA TGG TTT GAT GGA AGT AAC AAA AAC TAT GCA GAC TCC GTG 192 Ala Gly Ile Typ Phe Asp Gly Ser Asn Lys Asn Tyr Ala Asp Ser Val AAG GGC CGA TTC ACC ATC TCC AGA GAC AAT TCC AAG AAC ACG CTG TTT Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Phe 70 CTG CAA CTG AAC AGC CTG AGA GAC GAG GAC ACG GCT GTG TAT TAT TGT 288 Leu Gln Leu Asn Ser Leu Arg Asp Glu Asp Thr Ala Val Tyr Tyr Cys 85 GCG AGA GAG CGA GCA CGT GGT ATT TCT AGG TTC TAT TAC TAC ATG .Ala Arg Glu Arg Ala Ala Arg Gly Ile Ser Arg Phe Tyr Tyr Met 100 105 GAC GTC TGG GGC AAA GGG ACC ACG GTC ACC GTC TCC CCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Pro

120

- (2) INFORMATION FOR SEQ ID NO: 4 (LD1-40-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD1
- (B) CLONE: LD1-40
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: 2p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO.4 (LD1-40-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGC GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC ATT AGG AGC CAT TTG AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Arg Ser His Leu Asn TGG TAT CAG CAG AAA CCA GGG AAA GCC CCT AAG TTG CTG ATC TAT GGT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr Gly 40 GCG TCC ACT TTG CAA AGT GGC GTC CCA TCA AGG TTC AGT GGC AGT GGC Ala Ser Thr Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 50 55 TCT GGG GCA GTT TTC ACT CTC ACC ATC GCC AGT CTA CAA CCT GAA GAT Ser Gly Ala Val Phe Thr Leu Thr Ile Ala Ser Leu Gln Pro Glu Asp 70 80 TTT GCA ACT TAC TAC TGT CAA GAG AGT TAC AGT AAT CCT CTA ATC ACC Phe Ala Thr Tyr Tyr Cys Gln Glu Ser Tyr Ser Asn Pro Leu Ile Thr 85 TTC GGC CAA GGG ACA CGA CTG GAG ACT AAA 318 Phe Gly Gln Gly Thr Arg Leu Glu Thr Lys 100

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- (2) INFORMATION FOR SEQ ID NO: 5 (LD1-52-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL:LINE Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-52
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO.5 (LD1-52-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG Gln Val Lys Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GAA GCG TCT GGA TTC GCC CTC AGA AGT TCT Ser Leu Arg Leu Ser Cys Glu Ala Ser Gly Phe Ala Leu Arg Ser Ser GGA ATG CAC TGG GTC CGC CAG GCT CCT GGC AAG GGG CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 40 GCA CTT ATA TGG TTT GAT GGA AGT ATC AGA TCG TAT GCA GAA TCC GTG 192 Ala Leu Ile Typ Phe Asp Gly Ser Ile Arg Ser Tyr Ala Glu Ser Val 55 AAG GGC CGA TTC ACC ATC TCC AGA GAC ACT TCC AAG AAC ACC CTA TAT 240 Lys Gly Arg Phe Thr Ile Ser Arg Asp Thr Ser Lys Asn Thr Leu Tyr 70 75 CTC CAA ATG CGC AGT CTG AGT GCC GAC GAC ACG GCT GTG TAT TAC TGT Leu Gln Met Arg Ser Leu Ser Ala Asp Asp Thr Ala Val Tyr Tyr Cys GCG AGA GAC AAG GCG GTT CGG GGA ATT AGC AGG TAC AAC TAT TAC ATG Ala Arg Asp Lys Ala Val Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Met 110 GAC GTC TGG GGC AAA GGG ACC ACG GTC ACC GTC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser · 115 120

- (2) INFORMATION FOR SEQ ID NO: 6 (LD1-52-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-52
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO.6 (LD1-52-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AAC ATT ATC CGC TAT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Asn Ile Ile Arg Tyr Leu Asn 25 TGG TAT CAG CAG AAG CCA GGG AAA GCC CCT AGG CTC CTG ATC TAT GGT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Arg Leu Leu Ile Tyr Gly 40 GCG TCC ACT TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA 192 Ala Ser Thr Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 55 50 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGT AGT CTG CAA CCT GAA GAT 240 Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 65 TTT GCA ACT TAC TGT CAA CAG AGT TAC CGT ACC CCT CCA TTC ACT 288 Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Arg Thr Pro Pro Phe Thr 85 90 318 TTC GGC CCT GGG ACC AAA GTG GAG ATC AAA Phe Gly Pro Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO:7 (LD1-84-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-84
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION:q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO.7 (LD1-84-VH) CAG GTA AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG Gln Val Lys Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GAA GCG TCT GGA TTC ACC CTC AGA AGT TCT Ser Leu Arg Leu Ser Cys Glu Ala Ser Gly Phe Thr Leu Arg Ser Ser GGC ATG CAC TGG GTC CGC CAG GCT CCT GGC AAG GGG CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val ACA CTT ATA TGG TTT GAT GGA AGT ATC AGA TCG TAT GCA GAA TCC GTG Thr Leu Ile Typ Phe Asp Gly Ser Ile Arg Ser Tyr Ala Glu Ser Val 50 AAG GGC CGA TTC ACC ATC TCC AGA GAC ACT TCC AAG AAC ACC CTA TAT Lys Gly Arq Phe Thr Ile Ser Arg Asp Thr Ser Lys Asn Thr Leu Tyr 70 65 CTC CAA ATG CGC AGT CTG AGT GCC GAC GAC ACG GCT GTG TAT TAC TGT Leu Gln Met Arg Ser Leu Ser Ala Asp Asp Thr Ala Val Tyr Tyr Cys 90 85 GCG AGA GAC AAG GCG GTT CGG GGA ATT AGC AGG TAC AAC TAT TAC ATG 336 Ala Arg Asp Lys Ala Val Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Met 105 100 GAC GTC TGG GGC AAA GGG ACC ACG GTC ACC GTC TCC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 120

- (2) INFORMATION FOR SEQ ID NO: 8 (LD1-84-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-84
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 8 (LD1-84-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT ATA GGA Phe Thr Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Ile Gly Asp Arg GTC ACC ATC ACC TGC CGG GCA AGT CAG AGT ATC ATC AGG TAT TTG AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Ile Arg Tyr Leu Asn 25 TGG TAT CAG CAC AAA CCA GGA AAA GCC CCT AAA CTC CTC ATC TTT GCT Typ Tyr Gln His Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Phe Ala 40 GCA TCG AAT TTG CAA ACT GGG GTC CCA TCC AGG TTC AGT GGC AGT GGA 192 Ala Ser Asn Leu Gln Thr Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGT GAC CTG CAG CCT GAG GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Asp Leu Gln Pro Glu Asp 70 TTC GCA ACT TAC TAC TGT CAA CAG AGT TAC AGT AGG CCG TTC ACT TTT Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Ser Arg Pro Phe Thr Phe 85 90 GGC CGG GGG ACC AGC CTG GAC ATC AAA 315 Gly Arg Gly Thr Ser Leu Asp Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 9 (LD1-98-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-98
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 9 (LD1-98-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG 48 Gln Val Lys Leu Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GAA GCG TCT GGA TTC GCC CTC AGA AGT TCT Ser Leu Arg Leu Ser Cys Glu Ala Ser Gly Phe Ala Leu Arg Ser Ser 25 GAC ATA CAC TGG GTC CGC CAG GCT CCT GGC AAG GGG CTG GAG TGG GTG Asp Ile His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 40 GCA CTT ATA TGG TTT GAT GGA AGT ATC AGA TCG TAT GCA GAA TCC GTG 192 Ala Leu Ile Typ Phe Asp Gly Ser Ile Arg Ser Tyr Ala Glu Ser Val 55 AAG GGC CGA TTC ACC ATC TCC AGA GAC ACT TCC AAG AAC ACC CTA TAT Lys Gly Arg Phe Thr Ile Ser Arg Asp Thr Ser Lys Asn Thr Leu Tyr CTC CAA ATG CGC AGT CTG AGT GCC GAC GAC ACG CGT GTG TAT TAC TGT Leu Gln Met Arg Ser Leu Ser Ala Asp Asp Thr Ala Val Tyr Tyr Cys 90 GCG AGA GAC AAG GCG GTT CGG GGA ATT AGC AGG TAC AAC TAT TAC ATG 336 . Ala Arg Asp Lys Ala Val Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Met 100 GAC GTC TGG GGC AAA GGG ACC ACG GTC ACC GTC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 115

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- (2) INFORMATION FOR SEQ ID NO: 10 (LD1-98-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL:LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-98
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 10 (LD1-98-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA 48 Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 1.0 GTC ACC ATC ACT TGC CGG GCA AGT CAG AAC ATT ATC CGC TAT TTA AAT 96 Val Thr Ile Thr Cys Arg Ala Ser Gln Asn Ile Ile Arg Tyr Leu Asn 20 30 TGG TAT CAG CAG AAG CCA GGG AAA GCC CCT AGG CTC CTG ATC TAT GGT 144 Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Arg Leu Leu Ile Tyr Gly 40 GCG TCC ACT TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA 192 Ala Ser Thr Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGT AGT CTG CAA CCT GAA GAT 240 Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 TTT GCA ACT TAC TGT CAA CAG AGT TAC CGT ACC CCT CCA TTC ACT Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Arg Thr Pro Pro Phe Thr 95 TTC GGC CCT GGG ACC AAA GTG GAG ATC AAA 318 Phe Gly Pro Gly Thr Lys Val Glu Ile Lys

- (2) INFORMATION FOR SEQ ID NO: 11 (LD1-110-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-110
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 11 (LD1-110-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCT GGG AGG 48 Gln Val Lys Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Arg 10 TCC CTG AGA CTC TCC TGT ATA GCG TCT GGA TTC ACC CTC AGG AAT TAT 96 Ser Leu Arg Leu Ser Cys Ile Ala Ser Gly Phe Thr Leu Arg Asn Tyr 25 GCC ATG CAC TGG GTC CGC CAG GCT CCA GGA AAG GGG CTG GAG TGG GTG Ala Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 40 GCA GGT ATA TGG TTT GAT GGA AGC AAC AAA AAC TAT GCA GAC TCC GTG Ala Gly Ile Typ Phe Asp Gly Ser Asn Lys Asn Tyr Ala Asp Ser Val 55 AAG GGC CGA TTC ACC ATC TCC AGA GAC AAC TCC AAG AAC ACT CTG TTT Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Phe CTG CAC ATG AAC AGC CTG AGA GCC GAG GAC ACG GCT ACA TAT TAC TGT Leu His Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Thr Tyr Tyr Cys GCG AGA GAG AGG GCG ATT CGG GGA ATC AGT AGA TAC AAT TAC TAC ATG Ala Arg Glu Arg Ala Ile Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Met 100 GAC GTC TGG GGC AAG GGG ACC ACG GTC ACC GTC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 115 120

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- (2) INFORMATION FOR SEQ ID NO: 12 (LD1-110-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-110
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 12 (LD1-110-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC ATT CGA AGC TCT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Arg Ser Ser Leu Asn TGG TAT CAG CAG AAA CCA GGG AAA GCC CCT AAA GTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Val Leu Ile Tyr Ala GCA TCC AGT TTG CAA AGT GGG GTC CCA TCC AGG TTC AGT GGC AGA GGA Ala Ser Ser Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Arg Gly 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGC AGT CTG CAG CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 TTT GCG ACT TAT TAT TGT CAA CAG AGT TCC AGT TCC TCG TGG ACG TTC Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Ser Ser Ser Typ Thr Phe 85 GGC CAA GGG ACC AAG GTG GAA ATC AAA 315 Gly Gln Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 13 (LD1-117-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 378 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL: LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-117
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..345)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 13 (LD1-117-VH)

						TCA Ser										48
						GCA Ala										96
						CAG Gln										144
						GGC Gly 55										192
AAG Lys 65	GGC Gly	CGT Arg	TTC Phe	ACC Thr	ATC Ile 70	ÀCC Thr	AGA Arg	GAC Asp	AAC Asn	TCC Ser 75	AAG Lys	AAC Asn	ACG Thr	CTG Leu	TAT Tyr 80	240
						AGA Arg										288
GCG Ala	AGA Arg	GAG Glu	ACC Thr 100	TCA Ser	GTA Val	AGG Arg	CTA Leu	GGG Gly 105	TAT Tyr	AGC Ser	CGC Arg	TAC Tyr	AAT Asn 110	TAC Tyr	TAC Tyr	336
						GGG Gly										378

- (2) INFORMATION FOR SEQ ID NO: 14 (LD1-117-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE:-Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-117
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 14 (LD1-117 VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC ATT AGG AGC CAT TTG AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Arg Ser His Leu Asn TGG TAT CAG CAG AAA CCA GGG AAA GCC CCT AAG CTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr Ala 40 GCA TCC AGT TTG CAA GGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA Ala Ser Ser Leu Gln Gly Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 50 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGC AGT CTG CAA CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 80 75 65 70 TTT GCA ACT TAT TAC TGT CAA CAG AGT TAC AGG GCC CCT CAG TGG ACG Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Arg Ala Pro Gln Typ Thr 90 85 TTC GGC CAA GGG ACC AAG GTG GAA ATC AAA 318 Phe Gly Gln Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 15 (LD2-1-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE -- Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-1
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 15 (LD2-1-VH)

CAG Gln	GTG Val	AAA Lys	CTG Leu	CTC Leu 5	GAG Glu	TCT Ser	GGG Gly	GGA Gly	GGC Gly 10	GTG Val	GTC Val	C A G Gln	CCG Pro	GGG Gly 15	GGG Gly	48
TCC Ser	CTG Leu	AGA Arg	CTC Leu 20	TCC Ser	TGT Cys	GTA Val	GCG Ala	TCT Ser 25	GGA Gly	TTC Phe	ACC Thr	CTC Leu	AGG Arg 30	AGT Ser	TAT Tyr	96
GGC Gly	ATG Met	CAC His 35	TGG Typ	GTC Val	CGC Arg	CAG Gln	GCT Ala 40	CCA Pro	GGA Gly	AAG Lys	GGC Gly	CTG Leu 45	GAG Glu	TGG Typ	GTG Val	144
GCT Ala	TTT Phe 50	ATA Ile	TGG Typ	TTT Phe	GAT Asp	GGA Gly 55	AGT Ser	AAT Asn	AAA Lys	GGA Gly	TAT Tyr 60	GTA Val	GAC Asp	TCC Ser	GTG Val	192
AAG Lys 65	GGC Gly	CGA Arg	TTC Phe	ACC Thr	ATC Ile 70	TCC Ser	CGA Arg	GAC Asp	AAT Asn	TCC Ser 75	AAG Lys	AAC Asn	ATG Met	GTC Val	TAT Tyr 80	240
CTC Leu	CAA Gln	ATG Met	AAC Asn	AGC Ser 85	CTG Leu	AGA Arg	GCC Ala	GAT Asp	GAC Asp 90	ACG Thr	GCT Ala	GTA Val	TAT Tyr	TAT Tyr 95	TGT Cys	288
GCG Ala	AGA Arg	GAG Glu	AAG Lys 100	GCG Ala	CTT Leu	CGG Arg	GGA Gly	ATC Ile 105	AGC Ser	AGA Arg	TAC Tyr	AAC Asn	TAT Tyr 110	TAC Tyr	CTG Leu	336
GAC Asp	GTC Val	TGG Typ 115	GGC Gly	AAG Lys	GGG Gly	Thr	ACG Thr 120	GTC Val	ACC Thr	GTC Val	TCC Ser	TCA Ser 125				375

- (2) INFORMATION FOR SEQ ID NO: 16 (LD2-1-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 333 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE \leftarrow Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-1
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 22
- (B) MAP POSITION: q11.2
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (61..99, 145..165 , 262..294)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 16 (LD2-1-VL) GTG GTG ACT CAG CCA CCC TCA GCG TCT GGG ACC CCC GGA CAG AGG GTC Val Val Thr Gln Pro Pro Ser Ala Ser Gly Thr Pro Gly Gln Arg Val 10 ACC ATC TCT TGT TCT GGA AGC AAC TCC ATC CTT GGA AGT AAG TAT GTA Thr Ile Ser Cys Ser Gly Ser Asn Ser Ile Leu Gly Ser Lys Tyr Val 25 TAC TGG TAC CAG AAA CTC CCA GGA ACG GCC CCC AAA CTC CTC ATC TAT Tyr Typ Tyr Gln Lys Leu Pro Gly Thr Ala Pro Lys Leu Leu Ile Tyr 40 AAG AAT GAT CAG CGG CCC TCA GGG GTC TCT GAC CGA TTC TCT GGC TCC Lys Asn Asp Gln Arg Pro Ser Gly Val Ser Asp Arg Phe Ser Gly Ser 50 AAG TCT GGC ACC TCG GCC TCC CTG GCC ATC AGT GGG CTC CGG TCC GAG Lys Ser Gly Thr Ser Ala Ser Leu Ala Ile Ser Gly Leu Arg Ser Glu 65 70 80 GAT GAG GCT GAC TAT TAC TGT GCA CCA TGG GAT GCC AAC CTG GGT GGC 288 Asp Glu Ala Asp Tyr Tyr Cys Ala Pro Typ Asp Ala Asn Leu Gly Gly 85 90 CCG GTG TTC GGC GGA GGG ACC AAG CTG ACC GTC CTA AGT CAG CCC 333 Pro Val Phe Gly Gly Gly Thr Lys Leu Thr Val Leu Ser Gln Pro 105

- (2) INFORMATION FOR SEQ ID NO: 17 (LD2-4-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE ... Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-4
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 17 (LD2-4-VH) CAG GTG AAA CTG CTC GAG TCG GGG GGA GGC GTG GTC CAG CCG GGG GGG 48 Gln Val Lys Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly TCC CTG AGA CTC TCC TGT GAA GCG TCT GGA TTC ACC CTC AGA AGT TCT Ser Leu Arg Leu Ser Cys Glu Ala Ser Gly Phe Thr Leu Arg Ser Ser GGC ATG CAC TGG GTC CGC CAG GCT CCT GGC AAG GGG CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 40 GCA CTT ATA TGG TTT GAT GGA AGT ATC AGA TCG TAT GCA GAA TCC GTG Ala Leu Ile Typ Phe Asp Gly Ser Ile Arg Ser Tyr Ala Glu Ser Val 55 AAG GGC CGA TTC ACC ATC TCC AGA GAC ACT TCC AAG AAC ACC CTA TAT Lys Gly Arg Phe Thr Ile Ser Arg Asp Thr Ser Lys Asn Thr Leu Tyr 65 70 CTC CAA ATG CGC AGT CTG AGT GCC GAC GAC ACG GCT GTG TAT TAC TGT Leu Gln Met Arg Ser Leu Ser Ala Asp Asp Thr Ala Val Tyr Tyr Cys 85 90 GCG AGA GAC AAG GCG GTT CGG GGA ATT AGC AGG TAC AAC TAT TAC ATG Ala Arg Asp Lys Ala Val Arg Gly Ile Ser Arg Tyr Asn Tyr Met 100 GAC GTC TGG GGC AAA GGG ACC ACG GTC ACC GTC TCC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 115 120

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- (2) INFORMATION FOR SEQ ID NO: 18 (LD2-4-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 312 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-4
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..282)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(X1)	SEC	OENC	TE DI	SCR.	LPTIC	JN: S	SEQ .	TD NO). IE	3 (T-1)2-4.	- VL)		
				TCT Ser 5										48
				TGC Cys										96
				AAA Lys										144
				CAA Gln										192
				TTC Phe										240
				TAC Tyr 85									 -	 288
				GTG Val				:05						312

- (2) INFORMATION FOR SEQ ID NO: 19 (LD2-5-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-5
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 19 (LD2-5-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC TTG GTC CAG CCG GGG GGG Gln Val Lys Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT 96 Ser Leu Arg Leu Ser Cys Val Ala Ser Gly Phe Thr Phe Arg Ser Tyr GGA ATG CAC TGG GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val GCT TTT ATA TGG TTT GAT GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG Ala Phe Ile Typ Phe Asp Gly Ser Asn Lys Gly Tyr Val Asp Ser Val 50. AAG GGC CGA TTC ACC ATC TCC CGA GAC AAT TCC AAG AAC ATG CTC TAT Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Met Leu Tyr 70 CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC ACG GCT GTA TAT TAT TGT Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys 90 GCG AGA GAG AAG GCG CTT CGG GGA ATC AGT AGA TAC AAC TAT TAC CTG Ala Arg Glu Lys Ala Leu Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Leu 100 105 GAC GTC TGG GGC AAG GGG GCC ACG GTC ACC GTC TCA 375 Asp Val Typ Gly Lys Gly Ala Thr Val Thr Val Ser Ser 115 120

- 22) INFORMATION FOR SEQ ID NO: 20 (LD2-5-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-5
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:

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- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

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(D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 20 (LD2-5-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GTA TCT ATA GGC GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Val Ser Ile Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC GTT ACC AGG TCT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Val Thr Arg Ser Leu Asn TGG TAT CAG CAG AAA CCA GGG AAA GCC CCT AGG CTC CTA ATC TTT GGT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Arg Leu Leu Ile Phe Gly 35 40 GCG TCC ACT TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA 192 Ala Ser Thr Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly TCT GGG ACA GAT TTC ACC CTC ACC ATC AGC AGT CTG CAA CCT GAG GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 75 TTT GGA ACT TAC TGT CAA CAG AAT TAC AGG ACC CCT CAG TGG ACG Phe Gly Thr Tyr Tyr Cys Gln Gln Asn Tyr Arg Thr Pro Gln Typ Thr 85 TTC GGC CAA GGG ACC AAG GTA GAA ATC AAA 318 Phe Gly Gln Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 21 (LD2-10-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 378 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-10
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:

- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 298..345)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 21 (LD2-10-VH)

	CTG Leu							48
	CTC Leu 20							96
	TGG Typ							144
	TGG T y p						 	192
	TTC Phe							240
	AAC Asn							288
	GAG Glu 100					 	 	336
	TGG Typ							378

- (2) INFORMATION FOR SEQ ID NO: 22 (LD2-10-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 333 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL:TYPE ... Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-10
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 22
- (B) MAP POSITION: q11.2
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (61..102, 148..168, 265..294)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 22 (LD2-10-VL) GTG GTG ACT CAG GAG CCC TCA CTG ACT GTG TCC CCA GGA GGG ACA GTC Val Val Thr Gln Glu Pro Ser Leu Thr Val Ser Pro Gly Gly Thr Val ACT CTC ACC TGT GCT TCC AGC ACT GGG GCA GTC ACC AGG GGT TAC TAT Thr Leu Thr Cys Ala Ser Ser Thr Gly Ala Val Thr Arg Gly Tyr Tyr 20 25 30 CCA AAC TGG TTC CAG CAG AAG CCT GGA CAA GCA CCC AGG GCA CTG ATT Pro Asn Typ Phe Gln Gln Lys Pro Gly Gln Ala Pro Arg Ala Leu Ile 35 TAT AGT ACA AAC AAA AAA CAC TCC TGG ACC CCT GCC CGG TTC TCA GGC Tyr Ser Thr Asn Lys Lys His Ser Typ Thr Pro Ala Arg Phe Ser Gly TCC CTC CTT GGG GGC AAA GCT GCC CTG ACA CTG TCA GGT GTG CAG CCT Ser Leu Leu Gly Gly Lys Ala Ala Leu Thr Leu Ser Gly Val Gln Pro GAA GAC GAG GCT GAA TAT TAC TGC CTG CTC TAC TAT GGT GGT GCT CAA Glu Asp Glu Ala Glu Tyr Tyr Cys Leu Leu Tyr Tyr Gly Gly Ala Gln CTC GTA TTC GGC GGA GGG ACC AAG CTG ACC GTC CTA CGT CAG CCC Leu Val Phe Gly Gly Gly Thr Lys Leu Thr Val Leu Arg Gln Pro 100 105

- (2) INFORMATION FOR SEQ ID NO: 23 (LD2-11-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE -- Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-11
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:

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- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi)	SEÇ	QUENC	E DE	ESCRI	PTIC	ON: S	SEQ 1	D NC). 23	(LI	02-11	L - VH)	-		
				CTC Leu 5											48
				TCC Ser											96
				GTC Val											144
				TTT Phe											192
				ACC Thr											240
				AGT Ser 85											288
				GCG Ala											336
				AAA Lys											375

- (2) INFORMATION FOR SEQ ID NO: 24 (LD2-11-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-11
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 2
- (B) MAP POSITION: pl2
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 24 (LD2-11-VL) GTG TTG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT ATA CGA GAC AGA Val Leu Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Ile Arg Asp Arg GTC ACC ATC ACT TGC CGG GCA AGT CAG AAC ATT GGC AGT TAT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Asn Ile Gly Ser Tyr Leu Asn 20 TGG TAT CAG CAC AAA CCA GGG ACA GCC CCT AAA CTC CTG ATC TAT GCT Typ Tyr Gln His Lys Pro Gly Thr Ala Pro Lys Leu Leu Ile Tyr Ala 35 GTA TCC GCT TTG CAA AGT GGG GTC CCA TCG AGG TTC AGT GGC AGT AGA 192 Val Ser Ala Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Arg 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGC AGT CTG CAA CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 TTT GCA ACT TAC TGT CAA CAG AGT TAC AGT CCC CCG TAC ACT TTC Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Ser Pro Pro Tyr Thr Phe 85 90 GGG CAG GGG ACC AAC CTG CAG ATC AAA 315 Gly Gln Gly Thr Asn Leu Gln Ile Lys 100 105

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- (2) INFORMATION FOR SEQ ID NO: 25 (LD2-14-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-14
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 25 (LD2-14-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG Gln Val Lys Leu Leu Glu Ser Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA GTC GCC TGT GTA GCG TCT GGA TTC ACC TCC AGG AAT TTT Ser Leu Arg Val Ala Cys Val Ala Ser Gly Phe Thr Ser Arg Asn Phe 20 GGC ATG CAC TGG GTC CGC CAG GCT CCA GGA AAG GGG CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 35 GTT TTT ATT TGG TTT GAT GCA AGT AAT AAA GGA TAT GGA GAC TCC GTT 192 Val Phe Ile Typ Phe Asp Ala Ser Asn Lys Gly Tyr Gly Asp Ser Val 50 55 AAG GGC CGA TTC ACC GTC TCC AGA GAC AAT TCC AAG AAC ACG CTC TAT 240 Lys Gly Arg Phe Thr Val Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr CTG CAA ATG AAC GGC CTG AGA GCC GAA GAC ACG GCT GTA TAT TAT TGT 288 Leu Gln Met Asn Gly Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys 90 GCG AGA GAG AAG GCG GTT CGG GGA ATT AGT AGA TAC AAC TAC TAC ATG 336 Ala Arg Glu Lys Ala Val Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Met 100 105 GAC GTC TGG GGC AAG GGG ACC ACG GTC ACC GTC TCC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser ٠. 115 120

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- (2) INFORMATION FOR SEQ ID NO: 26 (LD2-14-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-14
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:

- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

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(D) OTHER INFORMATION:

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(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 26 (LD2-14-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTG GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC ATT ATC AAC AAT TTA AAT Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Ile Asn Asn Leu Asn TGG TAT CAG CAG AAA CCA GGC AAA GCC CCT GAA CTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Glu Leu Leu Ile Tyr Ala GCA TCC AGT TTG CAA AGT GGG GTC CCT TCA AGG TTC CGT GGC AGT GGA Ala Ser Ser Leu Gln Ser Gly Val Pro Ser Arg Phe Arg Gly Ser Gly 50 TCT GGG AGA GAT TTC ACT CTC ACC GTC ACC AGT CTG CAA CCT GAA GAT Ser Gly Arg Asp Phe Thr Leu Thr Val Thr Ser Leu Gln Pro Glu Asp TTT GCA ACT TAC TGT CAA CAG AGT TAC AGT AAC CCT GTG GAC GTT Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Ser Asn Pro Val Asp Val 85 90 CGG CAA GGG ACC AAG GTG GAA ATC AAA 315 Arg Gln Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 27 (LD2-17-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-17
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:

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- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 27 (LD2-17-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG Gln Val Lys Leu Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TCC AGG AGT TAT Ser Leu Arg Leu Ser Cys Val Ala Ser Gly Phe Thr Ser Arg Ser Tyr 25 GGA ATG CAC TGG GTC CGC CAG GCT CCA GGC AAG GGC CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val GCT TTT ATA TGG TTT GAT GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG 192 Ala Phe Ile Typ Phe Asp Gly Ser Asn Lys Gly Tyr Val Asp Ser Val 50 . AAG GGC CGA TTC ACC ATC TCC CGA GAC AAT TCC AAG AAC ACG CTC TAT 240 Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr 65 70 CTG CAA ATG AAG AGC CTG AGA GCC GAG GAC ACG GCT GTA TAT TGT 288 Leu Gln Met Lys Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys GCG AGA GAG AAG GCG CTT CGG GGA ATC AGT AGA TAC AAC TAT TAC CTG 336 Ala Arg Glu Lys Ala Leu Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Leu 100 105 GAC GTC TGG GGC AAG GGG ACC ACG GTC ACC GTC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 115 120

- (2) INFORMATION FOR SEQ ID NO: 28 (LD2-17-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-17
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 28 (LD2-17-VL) GTG ATG ACC CAG TCT CCA TTC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Phe Ser Leu Ser Ala Ser Val Gly Asp Arg 10 GTC ACC ATC ACT TGC CGG GCA AGT CAG AAC ATT AGG AGT TTT TTA AGT 96 Val Thr Ile Thr Cys Arg Ala Ser Gln Asn Ile Arg Ser Phe Leu Ser 25 20 TGG TAT CAG CAG AAA CCA GGG ACA GCC CCT AAG CTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Thr Ala Pro Lys Leu Leu Ile Tyr Ala 35 GCA TCC AGG TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGG Ala Ser Arg Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 50 : 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGC ACT CTG CAA CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Thr Leu Gln Pro Glu Asp TTT GCG ACT TAC TAC TGT CAA CAG AGT TAC AGT GCC CCT TGG ACG TTC 288 Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Ser Ala Pro Typ Thr Phe 90 85 315 GGC CAA GGG ACC AAG CTG GAA ATC AAA Gly Gln Gly Thr Lys Leu Glu Ile Lys

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- (2) INFORMATION FOR SEQ ID NO: 29 (LD2-18-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-18
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

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(D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 29 (LD2-18-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC TTG GTC CAG CCG GGG GGG Gln Val Lys Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT Ser Leu Arg Leu Ser Cys Val Ala Ser Gly Phe Thr Phe Arg Ser Tyr 20 25 GGC ATG CAC TGG GTC CGC CAG GCT CCA GGC AAG GGC CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 35 40 GCT TTT ATA TGG TTT GAT GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG 192 Ala Phe Ile Typ Phe Asp Gly Ser Asn Lys Gly Tyr Val Asp Ser Val 50 55 AAG GGC CGA TTC ACC ATC TCC CGA GAC AAT TCC AAG AAC ATG CTC TAT Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Met Leu Tyr 75 65 70 CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC ACG GCT GTA TAT TAT TGT 288 Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys 85 GCG AGA GAG AAG GCG CTT CGG GGA ATC AGT AGA TAC AAC TAT TAC CTG 336 Ala Arq Glu Lys Ala Leu Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Leu 100 GAC GTC TGG GGC AAG GGG ACC ACG GTA ACC GTC TCC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 115 120

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- (2) INFORMATION FOR SEQ ID NO: 30 (LD2-18-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 318 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD2
- (B) CLONE: LD2-18
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..288)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 30 (LD2-18-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GTA TCT ATA GGG GAA AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Val Ser Ile Gly Glu Arg 10 GTC ACC ATC ACT TGC CGG GAA AGT CAG AGC GTT ACC AGG TCT TTA ATT 96 Val Thr Ile Thr Cys Arg Glu Ser Gln Ser Val Thr Arg Ser Leu Ile 20 25 TGG TTT CAG AAG AAA CCA GGG AAA GCC CCT AGG CTC CTA ATC TTT GTT Typ Phe Gln Lys Lys Pro Gly Lys Ala Pro Arg Leu Leu Ile Phe Val 35 40 GCG TCC ACT TGG AAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA 192 Ala Ser Thr Typ Lys Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 50 - ___ 55 TCT GGG ACA GAT TTC ACC CTC ACC ATC AGC AGT CTG CAA CCT GAG GAT 240 Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 75 TTT GGA ACT TAC TGT CAA CAG AAT TAC AGG ACC CCT CAG TGG ACG 288 Phe Gly Thr Tyr Tyr Cys Gln Gln Asn Tyr Arg Thr Pro Gln Typ Thr 90 85 TTC GGC CAA GGG ACC AAG GTA GAA ATC AAA 318 Phe Gly Gln Gly Thr Lys Val Glu Ile Lys 100

- (2) INFORMATION FOR SEQ ID NO: 31 (LD2-20-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 375 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-20
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..342)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 31 (LD2-20-VH) CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC GTG GTC CAG CCG GGG GGG 48 Gln Val Lys Leu Leu Glu Ser Gly Gly Gly Val Val Gln Pro Gly Gly 10 TCC CTG AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TCC AGG AGT TAT 96 Ser Leu Arg Leu Ser Cys Val Ala Ser Gly Phe Thr Ser Arg Ser Tyr 20 GGC ATG CAC TGG GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG Gly Met His Typ Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Typ Val 35 GCT TTT ATT TGG TTT GAT GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG Ala Phe Ile Typ Phe Asp Gly Ser Asn Lys Gly Tyr Val Asp Ser Val 55 50 AAG GGC CGA TTC ACC ATC TCC CGA GAC AAT TCC AAG AAC ACG CTC TAT Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr 75 65 CTG CAA ATG AAG AGC CTG AGA GCC GAG GAC ACG GCT GTA TAT TAT TGT 288 Leu Gln Met Lys Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys 90 GCG AGA GAG AAG GCG CTT CGG GGA ATC AGT AGA TAC AAC TAT TAC CTG 336 Ala Arq Glu Lys Ala Leu Arg Gly Ile Ser Arg Tyr Asn Tyr Tyr Leu 105 GAC GTC TGG GGC AAG GGG ACC ACG GTC ACC GTC TCC TCA 375 Asp Val Typ Gly Lys Gly Thr Thr Val Thr Val Ser Ser 120

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- (2) INFORMATION FOR SEQ ID NO: 32 (LD2-20-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: cDNA LIBRARY, LD2
- (B) CLONE: LD2-20
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: CHROMOSOME 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142..162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 32 (LD2-20-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg GTC ACC ATC ACT TGC CGG GCA AGT CAG AGC ATT AGC AGC TAT TTA AAT 96 Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Ser Ser Tyr Leu Asn 20 TGG TAT CAG CAG AAA CCA GGG AAA GCC CCT AAG CTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr Ala 35 GCA TCC AGT TTG CAA AGT GGG GTC CCA TCA AGG TTC AGT GGC AGT GGA 192 Ala Ser Ser Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 55 TCT GGG ACA GAT TTC ACT CTC ACC ATC AGC AGT CTG CAA CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 75 TTT GCA ACT TAC TGT CAA CAG AGT TAC AGT ACC CGA TTC ACT TTC Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Tyr Ser Thr Arg Phe Thr Phe 85 90 315 GGC CCT GGG ACC AAA GTG GAT ATC AAA Gly Pro Gly Thr Lys Val Asp Ile Lys 100

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- (2) INFORMATION FOR SEQ ID NO: 33 (LD1-6-17-VH)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 384 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-6-17
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 14
- (B) MAP POSITION: q32.3
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (91..105, 148..198, 295..351)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence

(D) OTHER INFORMATION:

(xi)	(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 33 (LD1-6-17-VH)															
CAG (GTG Val	AAA Lys	CTG Leu	CTC Leu 5	GAG Glu	TCT Ser	GGG Gly	GGA Gly	GGC Gly 10	GTG Val	GTC Val	CAG Gln	CCT Pro	GGG Gly 15	AGG Arg	48
TCC (CTG Leu	AGA Arg	CTT Leu 20	TCC Ser	TGT Cys	GCA Ala	GCG Ala	TCT Ser 25	GGA Gly	TTT Phe	ACC Thr	TTC Phe	AGT Ser 30	AGC Ser	TAT Tyr	96
GGA . Gly !																144
ACA (192
AAG Lys 65																240
CTA Leu																288
GCG Ala																336
TAC Tyr					TGG Typ											384

- (2) INFORMATION FOR SEQ ID NO: 34 (LD1-6-17-VL)
- (i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 315 Base Pairs
- (B) TYPE: Nucleic acid with corresponding protein
- (C) STRANDEDNESS: Single
- (D) TOPOLOGY: Linear
- (ii) MOLECULE TYPE: cDNA to mRNA
- (iii) HYPOTHETICAL: No
- (iv) ANTI-SENSE: No
- (vi) ORIGINAL SOURCE: N-terminal fragment
- (A) ORGANISM: Homo sapiens
- (B) STRAIN: Not applicable
- (C) INDIVIDUAL / ISOLATE: Hyperimmune Rhesus D donor
- (D) DEVELOPMENT STAGE: Adult, Rearranged development pattern
- (E) HAPLOTYPE: Diploid
- (F) TISSUE TYPE: Not applicable
- (G) CELL TYPE: Peripheral lymphocyte B
- (H) CELL LINE: Not applicable
- (I) ORGANELLE: Not applicable
- (vii) IMMEDIATE SOURCE: Escherichia coli
- (A) LIBRARY: CDNA LIBRARY, LD1
- (B) CLONE: LD1-6-17
- (viii) POSITION IN GENOME:
- (A) CHROMOSOME / SEGMENT: Chromosome 2
- (B) MAP POSITION: p12
- (C) UNITS: Chromosome band number
- (xi) FEATURE:
- (A) NAME / KEY: CDR1, CDR2, CDR3
- (B) LOCATION: join base pair (64..96, 142.-162, 259..285)
- (C) IDENTIFICATION METHOD: By similarity with a known sequence
- (D) OTHER INFORMATION:

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 34 (LD1-6-17-VL) GTG ATG ACC CAG TCT CCA TCC TCC CTG TCT GCA TCT GTA GGA GAC AGA Val Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg 10 5 GTC ACC ATC ACT TGC CGG GCA AGT CAG GGC ATT AGA AAT GAT TTA ACC 96 Val Thr Ile Thr Cys Arg Ala Ser Gln Gly Ile Arg Asn Asp Leu Thr 25 20 TGG TAT CAG CAA AAA CCA GGG AAA GCC CCT AAG CTC CTG ATC TAT GCT Typ Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr Ala 35 GCA TCC AAT TTA CAA AGT GGG GTC CCA TCA AGG TTC AGC GGC AGT GGA 192 Ala Ser Asn Leu Gln Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly 55 50 TCT GGC ACA GAT TTC ACT CTC ACC ATC AGC CTG CAG CCT GAA GAT Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp 70 TTT GCA ACT TAT TAC TGT CTA CAA GAT AAC AAT TTC CCG TAC ACT TTT Phe Ala Thr Tyr Tyr Cys Leu Gln Asp Asn Asn Phe Pro Tyr Thr Phe 90 85 315 GGC CAG GGG ACC AAG CTG GAG ATC AAA Gly Gln Gly Thr Lys Leu Glu Ile Lys 100

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Fig. 1a

LD1-28-VH sequence

	9					18			27			36			45			54
5 '	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	G	S	L
			6 3															
	707	CTIC	63		<i>C</i> 2 2 2		mam		81	~~~		90			99			108
	AGA		100	161	GAA		101	GGA	TTC	GCC	CTC	AGA	AGT	TCT	GGC	ATG	CAC	TGG
	Ŕ	L	S		E	· 2	 C	G		 7	т т				G			
	••		_	_	٠,-	7.	3	G	r	A	יו	K						W
			117			126			125			144			CDRI			
	GTC	CGC											CCN		ATA		T C C C C C C C C C C C C C C C C C C C	
						- - -				GAG			GCA	C11	AIA	166	111.	GAT
	ν	R	0	А	P	G	K	G		Ε			A	L	т	W	=	D
			-						• •	_	•••	·			_			D
			171			180			189			100			207	CDR2		216
	GGA	AGT													ACC	אידיר	TCC	
														-		AIC		AGA
	G	S	I	R	S	Y	А	E	S	А	K	G	R	F	Т	I	s	Ŕ
														_	_	_	_	
			225			234			243			252			261			270
	GAC	ACT	TCC	AAG	AAC										AGT	GCC	GAC	GAC
					-													
	- D	T	S	K	N	T	٤	Y	L	H	M	R	s	L	S	Α	D	D
	٠.																	
•			279			288			297			306			31,5		~	324
- 2-1	ACG	GAT	GTG	TIT	TAC	TGT	GCG						CGG	GGA	TTA	AAC	AGG	TAC
	T	D	V	F	Y	С	А	R					R		I		R	Y
													- CDF	२३ —				
	220		333	.											369			
	AAC	TAT	TAC	ATG	GAC	GTC	TGG	GTC	AAA	GGG	ACC	ACG	GTC	ACC	GTC	TCC	TCA	3 '
	n		Y	м		1,7	 t.:	v			-							
					ט	V	W	V	ĸ	G	T	Т	V	Ί΄	V	S	S	
•		(CDR3				•											

Fig. 1b

LD1-28-VL sequence

			9															54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
							- 											
	V	M	T	Q	S	P	S	S	L	S	Α	S	V	G	D	R	V	Т
			<i>c</i> ¬			72			01			9.0			99			108
	አ TCC	አ ርጥ	63 TCC					AAC										
	AIC	ACI																
	I	Т	 C	R	, A	S	Q	N	I	I	R	Y	L	N	W	Y	Q	Q
	_													→				
			117			126			135			144			153			162
	AAG	CCA	GGG	AAA	GCC	CCT	AGG	CTC	CTG	ATC	TAT	GGT	GCG	TCC	ACT	TTG	CAA	AGT
								- 										
	K	P	G	K	A	P	R	L	L	I	Y	G				L	_	S
												←			- CDR	-		
			171			180		000	189	CCN	mcm.	198	202		207		CTC	
	GGG	GTC	CCA	TCA	AGG	TTC	AGT	GGC	AGI	GGA	101		ACA	GAI	110	AC 1		
	G	v	P	S	R	F	S	G	S	G	S	G	Т	D	F	Т	L	Т
	G	V	F	3		•	J	Ü	Ū	J	•	_	_	_				
			225			234			243			252			261			270
	ATC	AGT	AGT	CTG	CAA	CCT	GAA	GAT	TTT	GCA	ACT	TAC	TAC	TGT	CAA	CAG	AGT	TAC
														- 				
	I	S	S	L	Q	P	E	D	F	Α	Т	Y	Y	С	Q	Q	S	Y
٠.						200			207			306			315			
•	ССТ	200	279	$CC\lambda$	ттс	288 200	$\tau \tau C$	GGC	297 CCT	GGG	ACC	200						
· 2-7 <u>—</u>									-									
	R	Т	P	P	F	T	F	G	P	G	T	K	V	E	I	K		
			CDR	3														

Fig. 2a

LD1-40-VH sequence

			9			18			27			36			45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCT	GGG	AGG	TCC	CTG
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	R	S	L
			63						0.7			0.0			0.0			7.00
	7 C 7	CTC	63	TOT	עידיע	72 CCC	TOT	CCA	81		CTC	90 AGG	ידית ת	ጥለጥ	99	አጥር	CNC	108
	AGA		1 C C	161	AIA		101		110	ACC		AGG	AAI	1M1		AIG	CAC	166
	R	L	s	С	ī	Α	S	G	F	т	L	R	N	Y	А	М	Н	W
	••	_	_	Ū	_	•-		•	-	-	_				- CDF			
			117			126			135			144	`		153		,	162
	GTC	CGC		GCT	CCA							GTG				TGG	TTT	
	V	R	Q	А	P	G	K	G	L	Ε	W	V	Α	G	I	W	F	D
																CDR2		
			171			180			189			198			207			216
	GGA	AGT	AAC	AAA	AAC	TAT	GCA	GAC	TCC	GTG	AAG	GGC	CGA	TTC	ACC	ATC	TCC	AGA
	G	S	N	K	N		A	D	S	V	K	G	R	F	Т	I	S	R
							₹2						•					
	~ ~ ~		225			234	ama	-		G. 3. 3.		252	200	CMC	261	0.00	CNC	270
	GAC	AAI	100	AAG	AAC	ACG		111		CAA	CIG	AAC	AGC		AGA	GAC	GAG	GAC
-	. D	N	S	К	N	T	L	F	L	0	L	N	S	L	R	D	E	D
	٠	• •	Ŭ	• • • • • • • • • • • • • • • • • • • •	••	•	-	•	-	×	_		-	_	••	_	~	=
•			279			288			297			306			315		•	324
٠ ٤٠ ـــ	ACG	GCT	GTG	TAT	TAT	TGT	GCG	AGA	GAG	CGA	GCA	GCA	CGT	GGT	ATT	TCT	AGG	TTC
	T	Α	V	Y	Y	С	Α	R	Ε	R	Α	Α	R	G	I	S	R	F
													- CD	R3 —				
						342			351			360			369			
			333															
	TAT	TAC		ATG	GAC			GGC	AAA	GGG	ACC	ACG	GTC	ACC	GTC	TCC	CCA	3'
			TAC			GTC	TGG											3 '
	TAT Y		TAC Y	ATG M CDR3	GAC D				AAA K	GGG G	ACC T	ACG T	GTC V	ACC T	GTC V		CCA P	3'

Fig. 2b

LD1-40-VL sequence

			9			18			27			36			45			54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGC	GAC	AGA	GTC	ACC
		- - -											- 					
	V	M	T	Q	S	P	S	S	L	S	A	S	V	G	D	R	V	T
			63			72			81			90	-		99			108
	ATC	ACT		CGG	GCA	AGT	CAG	AGC	TTA	AGG	AGC	CAT	TTG	AAT	TGG	TAT	CAG	CAG
		2																
	I	T	С	R	A	`S	Q	S	I	R	S	Н	L	N	W	Y	Q	Q
									CDR:	l —				\longrightarrow				
			117			126			135			144			153			162
	AAA	CCA	GGG	AAA	GCC	CCT	AAG	TTG	CTG	ATC	TAT	GGT	GCG	TCC	ACT	TTG	CAA	AGT
	K	P	G	K	Α	P	K	L	L	I	Y					L	_	S
															CDR2			\longrightarrow
																		216
	GGC	GTC	CCA	TCA	AGG	TTC	AGT	GGC	AGT	GGC	TCT	GGG	GCA	GTT	TTC	ACT	CTC	ACC
		-																
	G	V	P	S	R	F	S	G	S	G	S	G	A	V	F	Т	L	Т
			225			234			243			252			261			270
	ATC	GCC													CAA			
												- 	- 					
	·I	Α	S	L	Q	P	Ε	D	F	Α	T	Y	Y	С	Q	E	S	Y
	•					200			207			306			←			
:			279			288		~~~	297	000	N (" N	200	СТС	CNC	212			
· 2.5	- AGT	AAT	CCT	CTA	ATC	ACC	110		CAA		ACA						ر	
	s	N	P	L	I	Т	F	G	Q	G	Т	R	L	E	T	K		
		(CDR3				•											

Fig. 3a

LD1-52-VH sequence

5'	CAG	GTG	9 AAA	CTG	CTC	18 GAG						36 GTC					TCC	54 CTG
	Q	V	K	L	L	E	S	G	G	G	V	V	Q	P	G	G	S	L
			63			72			81			90	-		99			108
	AGA	CTC	TCC	TGT	GAA	GCG	TCT	GGA	TTC	GCC	CTC	AGA	AGT	TCT	GGA	ATG	CAC	TGG
	R	L	S	С	- E	A	s	G	F	A	L	R	s	S	G	М	Н	W
															CDR1			
			117			126			135			144			153			162
	GTC	CGC	CAG	GCT	CCT	GGC	AAG	GGG	CTG	GAG	TGG	GTG	GCA	CTT	ATA	TGG	TTT	GAT
	V	R	Q	А	P	G	K	G	L	E	W	V	A	L	I	W	F	D
														←		CDR2		
			171									198			207			216
	GGA	AGT	ATC	AGA	TCG	TAT	GCA	GAA	TCC	GTG	AAG	GGC	CGA	TTC	ACC	ATC	TCC	AGA
			- - -															 D
	G	S	I	R	S	Y	А	Ε	S	V	K	G	R	F	T	I	S	R
			225			- CDR			242						261			270
	CNC	3 CT	225	220	7 7 C	234	OT: N			C 2 2		252	N C TT	CTC.		ccc		270
	GAC	ACI	TCC	AAG	AAC	ACC	CIA	IAI		CAA	AIG		AG1		AGI		GAC	GAC
-	D	Т	S	K	N	T	L	Y	L	Q	M	R	S	L	S	Α	D	D
•																		
																		324
	ACG	GCT	GTG	TAT	TAC	TGT	GÇG	AGA	GAC	AAG	GCG	GTT	CGG	GGA	ATT	AGC	AGG	TAC
	T	 А	v	Y	Y	C	A	R	D	 v		v	 D		 T		R	Y
	1	А	٧	1	1	C	A	К									R	1
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	אאר	ייי איזי	333 TAC		GAC		TCC				אככ		GTC		369 GTC	TCC	TCA	3 '
	AAC	1A1	:AC	AIG	GAC				AAA			ACG						د
	N	Y	Y	М	D	v	W	G	K	G	T	Т	V	Т	V	S	S	
			CDR3				→											

Fig. 3b

LD1-52-VL sequence

			9			18			27			36			45			54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
	v	М	Т	Q	s	P	S	S	L	S	A	S	V	G	D	R	V	Т
			63			72			81			90	-		99			108
	ATC	ACT	TGC	CGG	GCA	AGT	CAG	AAC	TTA	ATC	CGC	TAT	TTA	AAT	TGG	TAT	CAG	CAG
		L										- 			-			
	I	T	С	R	·A	`S	Q	N	I	I	R	Y	L	N	W	Y	Q	Q
									CDR					 →				
						126			135			144						
	AAG	CCA	GGG	AAA	GCC	CCT	AGG	CTC	CTG	ATC	TAT	GGT	GCG	TCC	ACT	TTG	CAA	AGT
	K	P	G	K	Α	P	R	L	L	I	Y	G	Α	_	Т	L	Q	S
												\leftarrow			- CD	R2 —		
			171			180			189			198			207			216
	GGG	GTC	CCA	TCA	AGG	TTC	AGT	GGC	AGT	GGA	TCT	GGG	ACA	GAT	TTC	ACT	CTC	ACC
							-			-								
	G	V	P	S	R	F	S	G	S	G	S	G	Т	D	F	Т	L	T
												0.5.0			261			270
			225			234			243			252		mcm	201	CAC		
	ATC	AGT	AGT	CTG	CAA	CCT	GAA	GAT	TTT	GCA	AÇT.	TAC	TAC	161	ÇAA	CAG	AGI	IAC
_								D	 F	 А	 T		Y		0	0	s	Y
	- I	S	S	L	Q	Р	Ε	ט	r	A	1	1	•	C	, v	×		
	•		226			205			297			306			315	-	.,	
	ССТ	אככ	2/9	~~a	ጉ ተር	200	~ ↑(GGC	CCT	GGG	ACC				-			
	CG1	ACC															•	
	R	Т	P	P	F	T	F	G	P	G	T	K	v	E	I	K		
			- CDF	£3 —			>											

Fig. 4a

LD1-84-VH sequence

			9			18						36						54
5 '	CAG	GTA	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
						- 												
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	G	S	L
			63			72			0 1			9.0			99			108
	እርአ	СТС			GAA								AGT	TCT		ATG	CAC	
														-				
	R	L	s	С	Ε	A	S	G	F	T	L	R	S	S	G	M	Н	W
													←		CDR1			
			117															
	GTC	CGC	CAG	GCT	CCT	GGC	AAG	GGG	CTG	GAG	TGG	GTG	ACA	CTT	ATA	TGG	TTT	GAT
	V	R	Q	А	P	G	K	G	L	E	W	V		L	I	W	F	D
																CDR2		
			171										CCA		207	N TT C	TCC	216
	GGA	AGT	ATC	AGA	TCG	TAT	GCA	GAA	TCC	GTG	AAG	GGC	CGA	110	ACC	AIC	100	AGA
	G	S	I	R	S	 V	Δ	F	ς.	V	ĸ	G	R	F	Т	I	s	R
	G	٦	1		_	CDR2							,					
			225		,				243			252			261			270
	GAC	ACT			AAC							CGC	AGT	CTG	AGT	GCC	GAC	GAC
-																		
	. D	T	S	K	N	Ξ	L	Y	L	Q	M	R	S	L	S	Α	D	D
•	•											206			2.5			324
			279		TAC											NGC.		
	ACG	GCT	GTG	TAT	TAC		G C G	AGA	GAC						A11			
	T	A	v	Y	Y	_	A	R			А	v	R	G	I	S	R	Y
	•	7.	•	-	-	-								CDR3				
			333			342									369			
	AAC	TAT			GAC										GTC	TCC	TCA	3'
									-				- 					
	N	Y	Y	M	D	v	W	G	K	G	T	T	V	T	V	S	S	
			— CD	R3 —			→											

Fig. 4b

LD1-84-VL sequence

			9			18			27			36						54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	ATA	GGA	GAC	AGA	GTC	ACC
		- - -																
	V	M	T	Q	S	P	S	S	L	S	Α	S	I	G	D	R	V	T
															0.0			108
			63			72			81			90			99		C 2 C	
	ATC	ACC		CGG	GCA	AGT	CAG	AGT		ATC	AGG	TAT	TTG	TAA	166	TAT	CAG	CAC
														 N7	W	Y	0	Н
	Ι	Т	С	R	` A	S	-					Y		N	W	1	Q	11
				←														
			117			126			135			144			153			162
	AAA	CCA	GGA	AAA	GCC	CCT	AAA	CTC	CTC	ATC	TTT	GCT	GCA	TCG	AAT	TTG	CAA	ACT
				-					- 									
	K	P	G	K	Α	P	K	L	L	I	F	Α	Α	_	N		_	T
															CDR2			
			171															216
	GGG	GTC	CCA	TCC	AGG	TTC	AGT	GGC	AGT	GGA	TCT	GGG	ACA	GAT	TTC	ACT	CTC	ACC
	G	V	P	S	R	F	S	G	S	G	S	G	T	D	F	Т	L	T
												0.50			261			270
			225			234			243			252		mom		CNC		
	ATC	AGT	GAC	CTG	CAG	CCT	GAG	GAT	TTC	GCA	ACT	TAC	TAC	TGT	CAA	CAG	AGI	IAC
_	-								- 							0	S	Y
	I	S	D	L	Q	P	Ε	D	F	A	Т	Y	Y	С	Q	Q	3	1
	•					200			207			306			3 15			
•			279	mm.c	3 CT	∠ 8 8	ccc	ccc	237	N.C.C	λCC				-			
* 4- 	AGT	AGG	CCG	110	ACT	111	نانان	CGG	ىىي	ACC	AGC			ATC		_	•	
					т	F	G	R	G	T	9	Τ.	D	т	к			
	S	R	P	F	_	r	G	Т	G	1	ر			-	••			
		(CDR3															

Fig. 5a

LD1-98-VH sequence

			9			18						36			45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	G	S	L
			63			72			81			90			99			108
	ACA	CTC	TCC	TGT	GAA	GCG						AGA				ATA	CAC	TGG
		111					-							-			-	
	R	L	S	С	·Ε	Α	S	G	F	Α	L	R	S	S	D	I	Н	W
																		
			117			126			135			144						162
	GTC	CGC	CAG	GCT	CCT	GGC	AAG	GGG	CTG	GAG		GTG			ATA	TGG	T"I"I'	GAT.
	-														 I	 147	F	D
	V	R	Q	A	P	G	K	G	L	Ε	W	V		L				_
												100			207	CDR2		216
			171			180	003	C	189	CTC	אאכ	GGC						
	GGA	AGT	ATC	AGA	TCG	TAT	GCA	GAA	100	G16	AAG							
	 G	S	I	R	S	Y	A	Ε	s	v	K	G	R	F	T	I	S	R
						- CDF	₹2 —											
			225			234			243			252						
	GAC	ACT	TCC	AAG	AAC	ACC	CTA	TAT	CTC	CAA	ATG	CGC	AGT	CTG	AGT	GCC	GAC	GAC
-																	- - -	D
	. D	T	S	K	N	T	L	Y	L	Q	М	R	S	L	S	Α	D	ט
	•					200			207			306			315			324
 2 - 1		205	279		י ידארי	288 707	ccc	ΛCΔ	CAC	בעם	GCG	GTT						
	ACG	CGI	GIG	+ 4A1	IAC													
	T	A	v	Y	Y		А	R	D	K		v						Y
																		
			333	i		342			351			360						
	AAC	TAT	TAC	ATG	GAC	GTC	TGG	GGC	AAA	GGG	ACC	ACG	GTC	ACC	GTC	TCC	TCA	. 3'
		 V	·	 М	 D	 V	 W		 К	- G	 Т	T	v	Т	v	S	S	
	N		Y		_	٧.	**	3	• • •	•	-	-						
			— CL	R3 -														

Fig. 5b

LD1-98-VL sequence

			^			18			27			36			45			54
5'	GTG	ATG	9 ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
J	 V	 M	 T	Q	 S	 P	s	 S	 L	S	 А	s	v	G	D	R	v	Т
			63	000	GCA	72 NGT	CAG	ממ	81 ATT	ATC	CGC	90 TAT			99 TGG			108 CAG
	ATC 	ACT T	10C	 R							 R			 И	- W	Y	Q	Q
	-				GCC	126			CDR:	ı ——		144		TCC	153 ACT		CAA	162 AGT
	 K	 P	- G	 K	 А	P	 R		L		Y			S	T CDR2	L	Q	S →
	GGG	GTC	171 CCA	, TCA	AGG	180 TTC	AGT	GGC	189 AGT	GGA	TCT	198 GGG	ACA			ACT		216 ACC
	G	v	 P	S	R	F	s	G	S	G	S	G	T	D	F	T	L	Т
	ATC	AGT	225 AGT	CTG	; CAA	234 CCT	GAA	. GAT	243 TTT	GCA	ACT	252 TAC	TAC	TGT	261 CAA		AGT	270 TAC
-	. 	 S		L	Q	P	 Е	D	- - -	Α	T		Y	C	Q		S	Y
- 12/4	CGT	` ACC	279 CC3	e r cc <i>i</i>	A TTC	288 ACT	TTC	GGC	297 CCT	ggc	acc		GTG			AA.		
	 R	T	P		F	T	F	G	P	G	T	K	V	Е	I	K		
			- CD	R3 —			→											

Fig. 6a

LD1-110-VH sequence

			9			18			27			36			45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCT	GGG	AGG	TCC	CTG
	Q	v	K	L	L	E	s	G	G	G	V	v	Q	P	G	R	S	L
	AGA	€TC	63 TC C	TGT	ATA	72 GCG		GGA			CTC	90 AGG		TAT		ATG	CAC	108 TGG
	 R	 L	S	 С	 I	~ А	s	G	F	 T	 L	R	N	Y	A CDR1	M	н	W
	GTC	CGC	117 CAG	GCT	CCA	126 GGA	AAG			GAG		144 GTG	GCA		153		TTT	162 GAT
	 V	 R	Q	 А	 P	 G	 К	 G	L	E	 W				I	W	F	D
	201		171	777	220	180	CCA	C Λ C	189 TCC	стс	באב	198 GGC		←	207	- CDF ATC		216 AGA
	GGA	AGC				1A1												
	G	S	N	K	N	Y	Α	D	S	V	K	G	R	F	Т	I	S	R
						- CDF	-		243) 252	•		261			270
•	GAC	AAC	225 TCC	AAG	AAC	234 ACT		TTT			ATG	AAC						
•	D	N	S	K	N	T	L	F	L	H	М	N	S	L	R	Α	E	D
: :::=	ACG	GCT	279 ACA	TAT	TAC		GCG	AGA	297 GAG	AGG	GCG	TTA		GGA	ATC	AGT		324 TAC
	T	 А	 Т	 Y	Y	 С	Α	R	E	R	A		R	G	I	S	R	Y
			333			342			351			360			369			
	TAA	TAC	TAC	ATG	GAC	GTC	TGG	GGC	AAG	GGG	ACC	ACG	GTC	ACC	GTC		1 CA	. 3'
	N	Y	Y CDR3	м 	D	v	₩ >	G	К	G	Т	Т	v	Т	V	S	S	

Fig. 6b

LD1-110-VL sequence

			9			18			27						45			54
5 '	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
-							- 											
	V	M	T	Q	S	P	S	S	L	S	Α	S	V	G	D	R	V	Т
						7.0			Ω1			9.0			99			108
	_		63	000	663	7.2	CAC	AGC	יייייע ע דס	CGA	A GC	ДСД	тта	AAT	TGG	TAT	CAG	CAG
	ATC	ACT		CGG	GCA	AG I	CAG	AGC	A11									-
	т	 Т	 C	TP	 Δ	· s	0	s	I	R	s	s	L	N	W	Y	Q	Q
	_	•	·	,					CDRI					 →				
			117												153			162
	אאא	CCA	GGG	444	GCC	CCT	AAA	GTC	CTG	ATC	TAT				AGT	TTG	CAA	AGT
	AAA															-		
	к	P	G	K	А	P	K	V	L	I	Y	A	Α	S	S	L	Q	S
	•	•	•									←			- CD	R2 —		
			171			180			189			198						216
	GGG	GTC	CCA	TCC	AGG	TTC	AGT	GGC	AGA	GGA	TCT	GGG	ACA	GAT	TTC	ACT	CTC	ACC
							-											
	G	V	P	S	R	F	S	G	R	G	S	G	T	D	F	T	L	T
									243			252			261			270
			225			234	~	a . T	243	000	νст	ער ב באת	тат	тст	C D D			
	ATC	AGC	AGT	CTG	CAG	CC.	GAA	GAT	111		ACI	171						-
_						 P	E	D	F	A	Т	Y	Y	С	Q	Q	s	S
	I	S	S	L	Q	r	L	L	•	••	-	_			<u> </u>			
			270			255			29 7			306			315		· .	
	AGT	TCC	TCG	TGG	ACG	TTC	GGC	CAA	GGG	ACC	AAG				AAA !	. 3'		
											-							
	S	S	S	W	T	F	G	Q	G	Т	K	V	Ε	I	K			
		— C	DR3 -			•												

Fig. 7a

LD1-117-VH sequence

5'	CAG	GTG	9 AAA	CTG	CTC	18 GAG		GGA								AAG	TCC	54 CTG	
_																			
	Q	V	K	L	L	Ε	S	G ,	G	G	V	V	Q	P	G	K	S	L	
			63			72												108	
	AGA	ÇTT	TCC	TGT	GCA	GCG	TCT	GGA					AGC		GGN	ATG	CAC	TGG	
	R	L	S	C	A	Α	S	G	F	S	F	N		Н	G	М	Н	W	
															CDR1		 →		
	GTC	CGC	117 CAG	GCT	CCA	126 GGC		GGG					GCA			TGG	TTT	162 GAT	
						~	+	 G	 T	 E	 tat	 V	Δ		т т	 W	F	D	
	V	R	Q	A	P	G	Λ.	G	11	E	**	V							
			171			180			189			198				CD.(L		216	
	GGC	AGT				TAT	GCA	GAC	TCC	GTG	AAG	GGC				ATC	ACC	AGA	
	 G	- S	 N	 K	 Y	 Y	 А	D	 S	v	 К	 G	 R	 F	-	I	T	R	
						— С													
			225			234						252		C.T.C		666		270	
_	GAC	AAC	TCC	AAG	AAC	ACG	CTG	TAT	CIN	CAA	ATG	AAC	AGC		AGA		GAG	GAC	
	, D	N	s	K	N	Ţ	L	Y	L	Q	М		S	L	Ŕ	A	E	D	
•			279			288			297			306			315		•	324	
	ACG	GCT	GTC					AGA					AGG	CTA	GGG	TAT	AGC	CGC	
														 T		 Y		 R	
	Т	Α	V	Y	Y	С	A	ĸ	Ē	7		V				1	3		
			333			347			351				— CD					378	
	TAC	AAT																TCA	3
				-								-							
	Y	N	Y	Ý.	M	כ	V	W	A	K	G	Т	Т	V	T	I	S	S	

Fig. 7b

LD1-117-VL sequence

			9			18			27			36			45			54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
	 V	 M	- Т	Q	s	P	S	s	L	S	A	S	V	G	D	R	v	Т
	ATC	ACT	63 TGC	CGG	GCA	72 AGT	CAG	AGC				90 CAT			99 TGG			108 CAG
	 I	<u></u>	<u>-</u>			-	- Q	 s	ī	R	s	H	L	N	W	Y	Q	Q
	AAA	CCA	117 GGG	AAA	GCC	126 CCT		СТС	135			144		TCC			CAA	
	К	 P	G	 K	 А	P	K	L	L	I	Y	A	A		S CDR2		Q	G →
	GGG	GTC	171 CCA	TCA	AGG	180 TTC	AGT	GGC	189 AGT	GGA	TCT	198 GGG			207			216 ACC
	 G	 V	 P	S	- R	F	s	G	S	G	S	G	Т	D	F	Т	L	Т
	ATC	AGC	225 AGT	CTG	CAA	234 CCT	GAA	GAT	243 TTT	GCA	ACT	252 TAT		TGT			AGT	270 TAC
-		- <i>-</i> -	S	 L	Q	 P	- Е	D	F	A	Т	Y	Y	C	Q	Q	S	Y
. 2.% -	AGG	GCC	279 CCT	CAG	TGG	288 ACG	TTC	GGC	297 CAA	. GGG	ACC	306 AAG	GTG	GAA	315 ATC			
	 R	Α	P	_	 W	T	- - - F	G	Q	G	Т	K	V	E	I	К		
			CD:	R3 —			→											

Fig. 8a

LD2-1-VH sequence

5'	CNC	CTC	9 מממ		СТС				27 GGA			36 GTC	CAG				TCC	54 CTG	
ο.																			
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	G	S	L	
			63			_						90						108	
	AGA			TGT	GTA	GCG	TCT	GGA			CTC	AGG	AGT	TAT		ATG	CAC	TGG	
	R	L	S	C	Λ.	A	S	G	 F	T	L	R			G CDR1		Н	W	
			117			126		222		626			←		153			162	
	GTC	CGC	CAG	GCT	CCA	GGA	AAG	GGC	CIG	GAG	166	GTG		111	AIA			GA1	
	V	R	Q	А	P	G	K	G	L	E	W	V	Α					D	
									100			100			207			216	
	GGA	AGT	171 AAT	AAA	GGA							GGC							
	G	s	N	K				D	S	v	К	G	R	F	Т	I	S	R	
			 .										•		261			270	
	GAC	AAT	225 TCC		AAC	234 ATG		TAT		CAA		AAC		CTG			GAT		
_	 D	N	s	K	N	M	v	Y	L	Q	M	N	s	L	R	A	D	D	
÷ - 2-%	ACG	GCT	279 GTA		TAT			AGA	GAG			CTT				AGC		324 TAC	
	т	 A	 V	 Y	 Y	 C	 A	 R	 E		 А	 L	 R	G	I	s	R	Y	
	•	7.	•	-	•	_	•••	• •	-						·				
	AAC	ТАТ	333 TAC		GAC		TGG			GGG					369		TCA	3 '	
	 N	 Y	 Y	 L	 D	 V	 W				 T	т	 V	 T	 V	- S			
		_	CDR3	_		· →	**	G	**	0	•	•	•	•	•	-	-		

Fig. 8b

LD2-1-VL sequence

			9			18			27			36			45			54
5'	GTG	GTG	ACT	CAG	CCA	CCC	TCA	GCG	TCT	GGG	ACC	CCC	GGA	CAG	AGG	GTC	ACC	ATC
•																		
	V	V	T	Q	P	P	S	Α	S	G	Т	P	G	Q	R	V	Т	I
						77			Q 1			90	•		99			108
	тст	TOT	63	CCA	A GC	AAC	TCC	ATC	CTT	GGA	AGT	AAG	TAT	GTA	TAC	TGG	TAC	CAG
	101	1,61																
	S	С	S	G	Š	N	S	I	L	G	S	K	Y	V	Y	W	Y	Q
								<u> —</u> с	DR1									
			117			126			135						153	~~~	222	162
	AAA	CTC	CCA	GGA	ACG	GCC	CCC	AAA	CTC	CTC	ATC	TAT	AAG	AAT	GAT	CAG		
									 T	 T	- - -			N	D	0	R	P
	K	L	P	G	T	Α	P	κ,	14	1,1	1	1						
						300			100			198				CDRZ		216
		~~~	171	TOT	CAC	180	الت	TCT	GGC	TCC	AAG	TCT	GGC	ACC				CTG
	TCA	فافاق	GIC	1 ( 1	GAC													
	ς.	G	v	S	D			S		S	K	S	G	T	S	Α	S	L
		Ŭ		_														
	,		225			234			243			252					221	270
	GCC	ATC	AGT	GGG	CTC	CGG	TCC	GAG	GAT	GAG	GCT	GAC	TAT	TAC	TGT	GCA	. CCA	166
-	. <b></b> -							- <del>-</del> -			<b>-</b>	- <del>-</del> -			C		P	W
	. А	I	S	G	L	R	S	E	D	Ę.	A	D	•	•	·			
•			279			288			297			306			315	•	14	324
: - '-	CAT	GCC	AAC	CTG	GGT	GGC	CCG	GTG	TTC	GGC	GGA	GGG	ACC	AAG	CTG	ACC	GTC	CTA
	D	A	N	L	G	G	P	V	F	G	G	G	T	K	L	Т	V	L
				— C!	DR3 -				<b>→</b>									
			333															
	AGT	CAG	CCC	3 '														
	S	Q	P															

Fig. 9a

#### LD2-4-VH sequence

			9			18						36			45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCG	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
																		L
	Q	V	K	L	L	Ē	S	G	G	G	V	V	Q	P	G	G	S	יו
			63			72			81			90			99			108
	AGA	CTC	TCC	TGT	GAA		TCT	GGA				AGA	AGT	TCT	GGC	ATG	CAC	TGG
						,												
	R	L	S	C	` E	Α	S	G	F	T	L	R	S	S	G	M		W
													<del></del>		CDR1		<del></del>	
			117															162
	GTC	CGC	CAG	GCT	CCT	GGC	AAG	GGG		GAG		GTG		CTT		TGG	T.I.I.	GAT
																 1.7	F	D
	V	R	Q	A	P	G	K	G	L	E	W	V	A			W		
														<del></del>		CDR2		
			171										CON		207	אידיר	TCC	216
	GGA	AGT	ATC	AGA	TCG	TAT	GCA	GAA	TCC	GTG	AAG	GGC	CGA	110	ACC	AIC	100	AGA
	 G	S	 I	 R	S	Y	A	E	S	v	ĸ	G	R	F	Т	I	S	R
	G	3	1			- CDF				<u> </u>		<u>`</u>						
			225						243			252			261			270
	GAC	ACT		AAG	AAC	ACC	CTA	TAT	CTC	CAA	ATG	CGC		CTG	AGT	GCC	GAC	GAC
<del>-</del> .			- <b></b>															
•.	D	T	S	K	N	T	L	Y	L	Q	M	R	S	L	S	A	D	D
•			279			220			297			306			315			324
· 2.5_	A C G	GCT			ТАС							GTT						
	ACG																	
	T	A	v	Y	Y	С	A	R	D	K	A	V	R		I			Y
									<del></del>					R3 —				<del></del>
			333			342						360			369			
	AAC	TAT	TAC	ATG	GAC	GTC	TGG	GGC	AAA	GGG	ACC	ACG	GTC	ACC	GTC	TCC	TCA	3 '
	 N	 Y	 Y	M	D	v	W	G	K	G	T	Т	v	Т	V	S	S	
	-		— رn	R3 —			<b>→</b>											

Fig. 9b

#### LD2-4-VL sequence

			9			18			27			36			45			54
5 '	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
	 V	 M	T	Q	s	P	s	S	L	S	Α	s	٧	G	D	R	v	T
	ΔΤΟ	ACT	63 TGC	CGG	ACA	72 AGT	CAG	ACC	81 ATT	AGC	AGA	90 AAT	TTA	CAT	99 TGG		CAG	108 CAG
	 I	 T	C	R	 T	 S	Q	T	I	s	R	N	L		W	Y	Q	Q
	AAA	CCA	117 GGG	AAA	GCC	126 CCT			135			144 GCT		TCC	153 AGT	TTG	CAA	162 AGT
	 К	 P	 G	К	 А	P	K	L	L	I	Y	A	Т	s	S CDR2		Q	S <del>→</del>
	GGG	GTC	171 CCA	TCA	AGG	180 TTC	AGT	GGC	189 AGT	GGA	TCT	198 GGG	ACA		207 TTC		CTC	216 ACC
	G	v	P	S	R	F	S	G	S	G	S	G	Т	D	F	T	L	T
	ATC	AAT	225 AGT	CTA	CAA	234 . CCT	GAA	GAT	243 TTT	GCA	ACT	252 TAC	TAC	TGT	261 CAA		AGT	270 TAC
-	I			- <del>-</del> - L		 P	E	D	F	A	Т	Y		С	Q	Q	s	Y
: : 2:1	- ACT	ACC	279 CCT	TCG	TTC	288 : GGC	CAA	. GGG	297 ACC	AAG	GTG	306 GAA	ATC		315		4	
	 T	- <b>-</b> -	 Р	S				 G	T	K	V	E	I	K				
		— CI	DR3 -		<b>→</b>													

Fig. 10a

#### LD2-5-VH sequence

S   CAG GTG AAA CTG CTC GAG TCT GGG GGA GGC TTG GTC CAG CCG GGG GGG TCC CTG  Q				9			18			27			36			45			54
AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT GGA ATG CAC TGG  R L S C V A S G F T F R S Y G M H W  117 126 135 144 153 162  GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG GCT TTT ATA TGG TTT GAT  V R Q A P G K G L E W V A F I W F D  171 180 189 198 207 216  GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  CDR2  225 234 243 252 261 270  GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC	5 '	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	TTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT GGA ATG CAC TGG  R L S C V A S G F T F R S Y G M H W  117 126 135 144 153 162  GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG GCT TTT ATA TGG TTT GAT  V R Q A P G K G L E W V A F I W F D  171 180 189 198 207 216  GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  CDR2  225 234 243 252 261 270  GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC		<b>-</b>																	
AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT GGA ATG CAC TGG  R L S C V A S G F T F R S Y G M H W  117		Q	V	K	L	L	E	S	G	G	G	L	V	Q	P	G	G	S	L
AGA CTC TCC TGT GTA GCG TCT GGA TTC ACC TTC AGG AGT TAT GGA ATG CAC TGG  R L S C V A S G F T F R S Y G M H W  117																0.0			100
R L S C V A S G F T F R S Y G M H W  117																	אידיר	CNC	
R L S C V A S G F T F R S Y G M H W						GTA	GCG	TCT	GGA	TTC	ACC	TTC	AGG	AGI	IAI	GGA	AIG	CAC	166
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$															v		м	ㅂ	พ
GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG GCT TTT ATA TGG TTT GAT  V R Q A P G K G L E W V A F I W F D  171 180 189 198 207 216  GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  225 234 243 252 261 270  GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC		R	Ъ	S	C	. <b>V</b>	A	5	G	r	1	r	K						
GTC CGC CAG GCT CCA GGA AAG GGC CTG GAG TGG GTG GCT TTT ATA TGG TTT GAT  V R Q A P G K G L E W V A F I W F D  171 180 189 198 207 216  GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  225 234 243 252 261 270  GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC														<del></del>					
V       R       Q       A       P       G       K       G       L       E       W       V       A       F       I       W       F       D         CDR2         CDR2         CDR2         CDR2         CDR2         225       234       243       252       261       270         GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC																			
V       R       Q       A       P       G       K       G       L       E       W       V       A       F       I       W       F       D         CDR2         171       180       189       198       207       216         GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA         GAC S       N       K       G       Y       V       D       S       V       K       G       R       F       T       I       S       R         CDR2         225       234       243       252       261       270         GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC		GTC	CGC	CAG	GCT							TGG				AIA	166	111	GAI
GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  CDR2  CDR3  CDR2  CDR3  CDR4  CDR4  CDR5  CDR5												1.7				 +	 M		D
GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  CDR2  225		V	R	Q	А	P	G	K	G	Ъ	L	W	V						
GGA AGT AAT AAA GGA TAT GTA GAC TCC GTG AAG GGC CGA TTC ACC ATC TCC CGA  G S N K G Y V D S V K G R F T I S R  225															<del></del>				
G S N K G Y V D S V K G R F T I S R							180												
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		GGA	AGT	TAA	AAA			GTA											CGA
CDR2  225 234 243 252 261 270  GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC																			D
225 234 243 252 261 270 GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC		G	S												Г	1	Τ.	3	K
GAC AAT TCC AAG AAC ATG CTC TAT CTG CAA ATG AAT AGC CTG AGA GCC GAG GAC																263			370
		GAC	AAT	TCC	AAG	AAC		CTC	TAL	CIG	CAA	AIG	AAI	AGC		AGA		GAG	GAC
D N S K N M L Y L Q M N S L R A E D			NT.		 У			 T	 v	Τ.	0	м	N	S	τ.	Ŕ	Δ	E	D
		٠.	IN	3	Λ.	14	1.		•	ם	¥	1.1			_	••	••	_	_
£ 279 288 297 306 315 × 324				279			288			297			306			315		ς.	324
ACG GCT GTA TAT TAT TGT GCG AGA GAG AAG GCG CTT CGG GGA ATC AGT AGA TAC		ACG.	GCT																TAC
and del din in the let ded not did the tree tree tree tree tree tree tree																			
TAVYYCAREKALRGISRY		т	А	V	Y	Y	С	А	R	Ε	K	Α	L	R	G	I	S	R	Y
← CDR3 ←		•	• •		_	_	_												
333 342 351 360 369				223			347												
AAC TAT TAC CTG GAC GTC TGG GGC AAG GGG GCC ACG GTC ACC GTC TCA 3'		አአር	ידמיד			GAC												TCA	3 '
AAC TAT TAC CIG GAC GIC 100 GGC 1210 GGC GGG TIOC GIC GAT			171																
N Y Y L D V W G K G A T V T V S S		N	Y	Y	L	D	V	W	G	K	G	Α	T	V	T	V	S	S	
———— CDR3 ———→				_ CD	R3 —			<b>→</b>											

# Fig. 10b

#### LD2-5-VL sequence

			9			18			27			36			45			54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GTA	TCT	ATA	GGC	GAC	AGA	GTC	ACC
_																		
	V	М	Т	Q	S	P	S	S	L	S	V	S	I	G	D	R	V	T
			<i>c</i> 3			72			Ω 1			90			99			108
		3 CM	63	ccc	CCA	7 Z	CNG	AGC	ليلت	ארר	AGG	TCT	TTA	AAT		TAT	CAG	CAG
	ATC	ACI	100			AG1												
	 I	T	C	R	`. A	S	Q	s	V	T	R	s	L	N	W	Y	Q	Q
	-	-	•						CDR:	ı —				<del></del>				
			117	`		126			135						153			162
	AAA	CCA	GGG	AAA	GCC	CCT	AGG	CTC	CTA	ATC	TTT	GGT	GCG	TCC	ACT	TTG	CAA	AGT
							<b>-</b>											
	K	P	G	K	Α	P	R	L	L	I	F	G	A		Т	L	Q	S
												←—			- CD	₹2 —		<del></del>
			171			180			189			198			207		omc	216
	GGG	GTC	CCA	TCA	AGG	TTC	AGT	GGC	AGT	GGA	TCT	GGG	ACA	GAT	TTC	ACC	C1C	ACC
													 Т	D	F	T	L	T
	G	V	₽	S	R	F	S	G	S	G	S	G	1	ט	ŗ	1		•
			225			224			243			252			261			270
	N TT C	700	225	СТС	ر م م ص	234 CCT	GAG	GAT	TTT	GGA	ACT					CAG	TAA	TAC
_	AIC	AGC	AG 1															
	ī	s	S	L	0	P	Ε	D	F	G	T	Y	Y	С	Q	Q	N	Y
•		_			_										<del></del>			
=			279			288			297			306			315		٠	
	AGG	ACC	CCT	CAG	TGG	ACG	TTC	GGC	CAA	GGG	ACC	AAG	GTA	GAA	ATC	AAA	' د .	
											т	 К	v	- <del></del>	Ī			
	R	T	P	Q	W	T	F	G	Q	G	1	K	v	_	-	• • • • • • • • • • • • • • • • • • • •		
			(	י אחי			<b>→</b>											

## Fig. 11a

#### LD2-10-VH sequence

			9			18												54	
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG	
	Q	v	K	L	L	E	s	G	G	G	v	v	Q	P	G	G	S	L	
	AGA	CTC	63 TCC	TGT	GTA	72 GCG	TCT	GGA	81 TTC	ACC	CTC	90 AGG	AGT	TAT	99 GGC			108 TGG	
	 R	 L	S	C	Α	`A	s	G	 F	T	L	R			_		H →	W	
	GTC	CGC	117 CAG	GCT	CCA	126 GGC	AAG	GGC	135 CTG	GAG	TGG		•		153			162	
	 V		 Q					 G					 А	 F	I	w	F	D	
	CGA	аст	171 AAT	444	GGA	180 TAT	GTA	GAC	189 TCC	GTG	AAG	198 GGC		TTC	207			216 CGA	
		 S			 G	 Y	 V	D	 S	 V	 K	G	 R					 R	
•	GAC	AAT	225 TCC	AAG		234		TAT	243			252		CTG		GCC		270 GAC	
	. D	N	s	K	N	M	v	Y	L	Q	M	Ŋ	s	L	R	А	D	D	
٠ د د د	ACG	GCT	279 GTA	. TAT	TAT	288 TAT	TGT	GCG	297 AGA	. GAG	AAG	306 GCG	CTT	CGG	315 GGA		AGC		
	T	A	V	Y	Y	Y	C	A	R			A						R	
	TAC	AAC	3 Š S TAT	TAC	CTG	342 GAC	GTC	TGG	GGC	AAG	GGG	360 ACC	ACG		369			378 TCA	3
	Y	N	Y	Y	L	D	V	W		K			T	V	T	v	S	S	

Fig. 11b

#### LD2-10-VL sequence

			9			18						36			45			54
5 '	стс	GTG	ΔCT	CAG	GAG	CCC	TCA	CTG	ACT	GTG	TCC	CCA	GGA	GGG	ACA	GTC	ACT	CTC
5																		
	V	V	T	Q	Ε	P	s	L	T	V	S	P	G	G	T	V	T	L
			63			72			81			90			99			108
	ACC	тст	GCT	TCC	AGC	ACT	GGG	GCA	GTC	ACC	AGG	GGT	TAC	TAT	CCA	AAC	TGG	TTC
	Т	С	A	S	S	T	G	Α	V	T	R	G	Y	Y	P	N	W	F
			-													$\longrightarrow$		
			117			126			135			144						
	CAG	CAG	AAG	CCT	GGA	CAA	GCA	CCC	AGG	GCA	CTG	TTA	TAT	AGT	ACA	AAC	AAA	AAA
	Q	Q	K	P	G	Q	Α	P	R	Α	L	I	Y	S	Т	N	K	K
														-		CDR	2 —	
			171			180			189									216
	CAC	TCC	TGG	ACC	CCT	GCC	CGG	TTC	TCA	GGC	TCC	CTC	CTT	GGG	GGC	AAA	GCT	GCC
	Н	S	W	T	P	Α	R	F	S	G	S	L	L	G	G	K	Α	A
		<b>-</b>																270
			225			234			243			252						270
	CTG	ACA	CTG	TCA	GGT	GTG	CAG	CCT	GAA	GAC				TAT	TAC	TGC	CTG	CIC
																		 T
	L	T	L	S	G	V	Q	P	Ε	D	Ε	А	E	Y	Y	C	L	L
•												206			315		<del></del>	324
•			279			288			297	000	C C 3	306	N.C.C	770	2.T.D		СТС	
1254	TAC	TAT	GGT	GGT	GCT	CAA	CTC	GTA	TTC	GGC	GGA	666	ACC	AAG			GTC	
													T	ĸ	Ť.	т	v	L
	Y	Y	-				ب ا	V	r	G	G	G	1	10		•	·	_
				- CDF	₹3 —			<del></del>										
			333															
		CAG																
	R	Q	P															

Fig. 12a

## LD2-11-VH sequence

			9			18			27			36	•		45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCG	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
	Q	V	K	L	L	Ε	S	G	G	G	V	V	Q	P	G	G	S	L
															0.0			100
			63			_			-		ama			mom.	99	3 mc	an a	108
	AGA	CTC	TCC	TGT	GAA	GCG	TÇT	GGA	TTC	ACC	CTC	AGA	AGT	TCT	GGC	ATG	CAC	166
			==							<b>-</b>	 T	R	s	s	G	М	н	W
	R	r	S	С	·Ε	A	S	G	F	1	بد	ĸ					11	**
															CDR1		<del></del>	1.60
			117										<b>663</b>		153			162
	GTC	CGC	CAG	GCT	CCT		AAG							CII	ATA	166	111	GAI
																1.7	 F	D
	V	R	Q	А	P	G	K	G	L	Ε	W	V	A	L	I	W		_
																CDR2		
			171												207			216
	GGA	AGT	ATC	AGA	TCG	TAT	GCA	GAA	TCC	GTG	AAG	GGC	CGA	TTC	ACC	ATC	TCC	AGA
																 -		
	G	S	Ι	R								G	R	P.	1	1	5	R
												<del></del>	•					
			225			234						252			261	~~~		270
	GAC	ACT	TCC	AAG	AAC	ACC	CTA	TAT	CTC	CAA	ATG	CGC	AGT	CTG	AGT	GCC	GAC	GAC
								Y			 М	R		L	s	 А	D	D
	. D	T	5	K	N	-	L	1	L	Q	141	ĸ	3	11	3	А	ט	ט
•			279			288			297			306			3.15			324
•	N C G	CCT		TAT														
	Т	А	V	Y	Y	С	А	R	D	ĸ	А	V	R	G	I	s	R	Y
	•		·	-	-	-							— رى	R3				
			333			342			` 351			360			369			
	אאר	тдт		ATG	GAC									ACC		TCC	TCA	3 '
	N	Y	Y	M	D	V	W	G	K	G	Т	T	v	T	V	S	S	
			- CD	R3 —														

Fig. 12b

## LD2-11-VL sequence

		9			18			27			36			45			54
GTG	TTG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	ATA	CGA	GAC	AGA	GTC	ACC
 V		 Т	0	 S	 P	 S	s	 L	 S	 А	S	I	R	D	R	v	T
v	ני	•	-														
		63			72			81			90						108
ATC	ACT	TGC	CGG	GCA	AGT	CAG	AAC			AGT	TAT	TTA	AAT	TGG	TAT	CAG	CAC
 I	 Т	- <del></del> -	<b>-</b>	 А`	- <del>-</del> - S	Q	N	I		S	Y	L	N	W	Y	Q	Н
			<del></del>					CDR	ı —				<del></del>	•			
		117			126			135			144						
AAA	CCA	GGG	ACA	GCC	CCT	AAA	CTC	CTG	ATC	TAT	GCT	GTA	TCC	GCT	TTG	CAA	AGT
							 L	- <del>-</del> -						Δ	T.	0	S
K	P	G	T	A	Р	K	Ŀ	ъ	1	1	. A			– CD			
											100				R2		216
		171	<b>T</b>		180 TTC	x C T	ccc	183	אכא	ጥርጥ	GGG						
GGG	GTC	CCA	TCG	AGG	1 4 C	AGI		AG1									
G	V	P	S	R	F	S	G	S	R	S	G	T	D	F	T	L	Т
		225			234			243			252			261			270
ATC	AGC	AGT	CTG	CAA	CCT	GAA	GAT	TTT	GCA	ACT	TAC	TAC	TGT	CAA	CAG	AGT	TAC
<del>-</del>				- <b>-</b> -													Υ
·.I	S	S	L	Q	P.	Ε	D	F	Α	Ţ	Y	Y	C	Q	Q	S	1
•		279			288			207			306			315		٠,	
TAGT	ccc	279 CCG	TAC	ACT	TTC	GGG	CAG	GGG	ACC	AAC							
S	P	P	Y	T	F	G	Q	G	Т	N	L	Q	I	K			
	— CI	R3 -		<del></del>													

Fig. 13a

#### LD2-14-VH sequence

			9			18							a. a	000	45	ccc	TCC	54 CTC
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG		GGG			
	Q	v	K	L	L	E	S	G	G	G	v	v	Q	P	G	G	S	L
	AGA	GTC	63 GCC		GTA			GGA		ACC			TAA	TTT	99 GGC	ATG	CAC	108 TGG
													<b>-</b>					
	R	V	Α	С	V	Α	S	G	F	T	S	R	N		G	M	Н	W
			117			126			135			144						162
	GTC	CGC	CAG	GCT									GTT	,I,,I,,I,	ATT	166	111	GA1
	v	 R	Q	A	P	 G	к	G		E	W	v	V	F	I	W	F	D
			171			180			189						207			216
	GCA	AGT	TAA	AAA	GGA	TAT	GGA	GAC	TCC	G'I"I'	AAG		CGA	110	ACC			
	Α	s	N	К	G	Y				V			R	F	T	V	S	R
			225	220		234	R2 —		243	C A A		252 AAC		CTG	261 AGA		GAA	270 GAC
_	GAC	AAI	100	AAG	AAC	ACG											- <b>-</b> -	<b>-</b>
	D	N	S	K	N	T	L	Y	L	Q	М	N	G	L	R	A	E	D
<del></del> - 2-%	ACG	GCT	279 GTA	TAT	TAT	288 TGT	. GCG	AGA	297 GAG	AAG	GCG		CGG		315 ATT		AGA	324 TAC
	<del>-</del>			- <b></b>											 I		- <del>-</del> -	Y
	T	Α	V	Y	Y	С	A	R	E	K								
						242	2		<del>←</del>				— CL	-				
	DAA	TAC	333 CAT :		GAC	342 GTC	TGG	GGC	AAG	GGG	ACC	ACG	GTC		GTC		TCA	. 3'
	n	Y	Y	 М	D	v	W	G	K			Т		Т	v	S	s	
			CDR.	3 —		<del></del>												

Fig. 13b

#### LD2-14-VL sequence

			9			18			27			36			45			54
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTG	GGA	GAC	AGA	GTC	ACC
	V	M	T	Q	S	P	S	S	L	S	Α	S	V	G	D	R	V	T
			<b>63</b>			72			Ω 1			9.0	-		99			108
	አጥር	א כיידי	63 TGC	CGG	GCA	AGT	CAG	AGC	TTA	ATC	AAC	AAT	TTA	AAT	TGG	TAT	CAG	
	AIC	ACI	-100															
	I	Т	С	R	` A	S	Q	S	I	I	N	N	L	N	W	Y	Q	Q
				←—					CDR	ı —				<b></b> →				
			117			126			135			144						
	AAA	CCA	GGC	AAA	GCC	CCT	GAA	CTC	CTG	ATC	TAT	GCT	GCA	TCC	AGT	TTG	CAA	AGT
		<b>-</b>																
	K	P	G	K	A	P	Ε	L	L	I	Y	A						S
												<del></del>			- CD	R2 —		<del>→</del> 216
			171			180			189	car		198	202		207	λСТ	CTC	
	GGG	GTC	CCT	TCA	AGG	TTC	CGT	GGC	AGT	GGA	1 ( 1		AGA	GAI	110			
	G	v	 Р	S	R	 F	R	G	s	G	S	G	R	D	F	T	L	Т
	G	V	F	3	K			Ü		Ū	•	_						
			225			234			243			252			261			270
	GTC	ACC	AGT	CTG	CAA	CCT	GAA	GAT	TTT	GCA	ACT	TAC	TAC	TGT	CAA	CAG	AGT	TAC
-																		
	V	T	S	L	Q	P	Ε	D	F	A	T	Y	Y	С	Q	Q	S	Y
_						200			207			206			<del>&lt;</del>			
_	<b>3</b> OFF	220	279	GTG	CNC	288	CCC	CNN	297	» CC	ממ							
	AGT	AAC		G 1 G	GAC											-		
	S	N	P	v	D	v	R	Q	G	Т	K	v	E	I	K			
			- CDF	23		<b>→</b>												

÷

## Fig. 14a

#### LD2-17-VH sequence

			9			18			27			36			45			54
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
ر																		
	Q	V	K	L	L	Ē	S	G	G	G	V	V	Q	P	G	G	S	L
			<i>-</i>			72			81			90			99			108
•	202	CTC	63 TCC	тст	GTA	GCG	тст	GGA	TTC	ACC	TCC	AGG	AGT	TAT	GGA	ATG	CAC	TGG
	AGA		100			-,												
	R	L	S	С	V	A	S	G	F	T	S	R	S	Y	G	M	Н	W
													•		CDR1		<del></del>	
			117			126			135						153		$\tau \tau \tau$	162
	GTC	CGC	CAG	GCT	CCA	GGC					TGG	GTG	GC'I'	.111.	AIA	166	111	
									 L	 E	 W	 V	A	F	I	W	F	D
	V	R	Q	А	P	G	K	G	יר	E	**	•		-				
						100			189			198						216
		3 CM	171	א <i>א</i> א	GGA	TAT	ста	GAC	TCC	GTG	AAG	GGC	CGA	TTC	ACC	ATC	TCC	CGA
	GGA	AG1	AAI								<del>-</del>		<b>-</b>					
	G	S	N	K	G	Y	V	D	S	V	K	G	R	F	T	I	S	R
						- CD1	R2 —					;	<b>&gt;</b>					270
			225			234			243			252				000		
	GAC	TAA	TCC	AAG	AAC	ACG	CTC	TAT	CTG	CAA	ATG	AAG	AGC	CTG	AGA			
-	. <b></b> -								 T		 M	ĸ	s	L	R	Α	E	D
	-, D	N	S	K	N	Т	L	1	ב	Q	1.1		Ū	_				
-			279			288			297			306						
· 2.7_	acc	GCT	272 GTA	TAT	TAT	TGI	GCG	AGA	GAG	AAC	GCG	CTI	CGG	GGA	ATC	AGT	AGA	TAC
	T	Α	V	Y	Y	C	А	R	Ē	K						S	R	Y
									•					_				
			333	3		342	2		351			360	) . cæc	3 300	369	י דריר	י דרב	٠ , ,
	AAC	TAT	TAC	CTC	GAC	GTO	TGC	G GGC	AAC	GGC	ACC	ACC						3'
	 N	 Y	 Y	·	<del>-</del> D		 W	 G			Т	Т	v	Т	V		S	
		_	-	DR3 -	_		$\rightarrow$											

Fig. 14b

#### LD2-17-VL sequence

5'	GTG	ATG	9 ACC	CAG	TCT	18 CCA	TTC	TCC	27 CTG	TCT	GCA	36 TCT		GGA		AGA	GTC	54 ACC
	 V	 M	 T	 Q	S	P	 F	S	L	S	A	s	v	G	D	R	v	T
	ATC	ACT	63 TGC	CGG	GCA	72 AGT	CAG	AAC	81 ATT	AGG	AGT _.	90 TTT	ATT	AGT	99 TGG	TAT	CAG	108 CAG
	 I	 T		 R	 A	s ·	 Q	n			s			S	W	Y	Q	Q
	_			<del></del>					— CD	R1 -				<del></del>				
	AAA	CCA	117 GGG	ACA	GCC	126 CCT	AAG	CTC	135 CTG	ATC	TAT	144 GCT	GCA	TCC	153 AGG	TTG	CAA	162 AGT
															 R	L	0	S
	K	P	G	T	Α	P	K	L	L	I	Y	Α	A	S	DR2		V	
	000	CTC.	171	TCA	NGG	180 TTC	AGT	GGC	189 AGT	GGG	TCT	198 GGG			207		CTC	216 ACC
	ال ال	G I C											<del>-</del>					
	G	V	P	S	R	F	S	G	S	G	S	G	Т	D	F	Т	L	Т
	<b>Δ</b> Τ.(	AGC	225 ACT	· CTG	CAA	234 . CCT	GAA	GAT	243 TTT	· GCG	ACT	252 TAC	TAC	TGT	261 CAA		AGT	270 TAC
_																		Y
	Ţ	S	T	L	Q	P	Ξ	D	F	Α	Т	Y	Y	С	Q	Q	S	1
	AGT	GCC	279 CCT	TGG	ace	288 TTC	GGC	CAA	297 . GGG	ACC	: AAG		GAA		315 AAA		*	
	S	A	P	 W	T	F	 G	Q	G	T	K	L	E	I	K			

Fig. 15a

#### LD2-18-VH sequence

			9			18			27			36			45			54
5 '	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	TTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
_																		
	Q	V	K	L	L	Ε	S	G	G	G	L	V	Q	P	G	G	S	L
			63			72			81			90	-		99			108
	AGA	CTC	TCC	TGT	GTA	GCG	TCT	GGA	TTC	ACC	TTC	AGG	AGT	TAT	GGC	ATG	CAC	TGG
			-				<del>-</del>		<del>-</del>			- <b></b>						
	R	L	S	С	· V	A	S	G	F	T	F	R	S	Y	G	M	Н	W
													<del></del>		CDR1		<del></del>	
			117			126			135			144						162
	GTC	CGC	CAG	GCT	CCA	GGC	AAG	GGC	CTG	GAG	TGG	GTG	GCT	TTT	ATA	TGG	T"I"I	GAT
															- <del>-</del> -	 W	F	D
	V	R	Q	А	P	G	K	G	L	E	W	V			I		=	
												100			207	CDRZ		216
			171			180		~~~	189	CTC.	220					ΔΤС	тсс	
	GGA	AGT	TAA	AAA	GGA	TAT	GTA	GAC	TCC	GIG	AAG	GGC						
	G	S	N	K	G							G		F	T	I	S	R
							R2 —					<del></del>			261			270
			225			234			243			252		CTC		CCC		
	GAC	AAT	TCC	AAG	AAC	ATG	CTC	TAT	CTG	CAA	ATG	TAA	AGC	CIG	AGA		GAG	
<del>-</del> .	D	N	S	K	N	М	L	Y	L	Q	М	N	s	L	R	Α	E	D
			256			200			297			306			315			324
		~~	279	y	ت لابات	∠55 TCT	GCG	. מכם	CAG	AAG	GCG	CTT						
	ACG	GCI	المراجئ	. iAI	IAI													
	T	A	v	Y	Y		А	R				L			I			Y
														R3 -				
			3 3 3	1		342								N C C	369 CTC		тсъ	٦,
	AAC	TAT	TAC	CTG	GAC	GTC	TGG	, GGC	AAG	تانان د	ACC	ACG	GIA	. ACC				. 3'
	N	Y	Y		D	V	W	G	K	G	Т	Т	v	Т	V	S	S	
			— CI	R3 -			$\rightarrow$											

Fig. 15b

## LD2-18-VL sequence

			_			10			27			36			45			54
5'	GTG	ATG	9 ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GTA	TCT	ATA	GGG	GAA	AGA	GTC	ACC
	- <b>-</b> -	 M	 T		 S	 P	 S	 S	L		v	s	I	G	E	R	v	T
	·		63	_		72			81			90	٠		99		<b>63.6</b>	108
	ATC	ACT	TGC	CGG	GAA	AGT	CAG	AGC	GTT	ACC	AGG	TCT	TTA	ATT	TGG	1.1.1.	CAG	
	 I	 T	C	R	`E	S	Q				R	S		I	W	F	Q	K
			117	<del></del>		126			135			144		<del></del> >	153			162
	AAA	CCA	GGG	AAA	GCC	CCT	AGG	CTC	CTA	ATC	TTT	GTT	GCG	TCC	ACT	TGG	AAA 	AG 1
	 K	 P	- <i></i> G	 К	 А	 Р	 R		L	I	F	V	А	S			K	S
															2 —			<del>→</del> 216
	GGG	GTC	171 CCA	TCA	AGG	180 TTC	AGT	GGC	189 AGT	GGA	TCT	GGG	ACA	GAT				
		 V				 F						G						
			225			234			243			252	m > C	mca.			דממ:	
	ATC	AGC	AGT	CTG	CAA	CCT	GAG	GAT	TTI	' GGA	ACT	TAC	TAC	161		. CAC		
-		 S	 S	 L	Q	P	E	D	F	G	Т	Y	Y	С	Q	Q	N	Y
•	· · AGG	ACC	279 CCT	; CAG	; TGG	289 ACC	; ; TTC	: GGC	297 CAA	, A GGC	ACC	306 AAG	GTA	GA#			3'	
	 R	 T	 P		 W	T	F	 G	Q	 G	т		v	E		K	-	
			(	DR3			<b>→</b>											

Fig. 16a

#### LD2-20-VH sequence

			9			18			27			36						
5'	CAG	GTG	AAA	CTG	CTC	GAG	TCT	GGG	GGA	GGC	GTG	GTC	CAG	CCG	GGG	GGG	TCC	CTG
	<b>-</b>		<b>-</b>											 P		 G		L
	Q	V	K	L	L	E	S	G	G	G	V	V	Q	r	0	Ü	J	_
			63			72			81			90						108
	AGA	CTC	TCC	TGT	GTA	GCG	TCT	GGA	TTC	ACC	TCC	AGG	AGT	TAT	GGC	ATG	CAC	TGG
						- ,						 R						w
	R	L	S	С	V	Α	S	G	F.	1.	5	ĸ	<i>→</i>		CDR1			••
						176			1 3 5			144	<b>—</b>		153		ŕ	162
	GTC	CGC	117 CAG	GCT	CCA	GGA	AAG	GGC	CTG	GAG	TGG	GTG	GCT	TTT	TTA	TGG	TTT	GAT
	V	R	Q	А	P	G	K	G	L	E	W	V		F		W	_	D
												100		<del></del>				216
			171		~~~	180	GTA	C 1 C	189	CTC	אאכ	198						
	GGA	AGT	AAT	AAA	GGA	1A1	GIA	GAC										
	G	S	N	К	G	Y	V	D	S	V	K	G	R	F	T	I	S	R
						- CDI	R2 —					<b>→</b>						270
			225			234			243							000		270 GAC
_	GAC	TAA	TCC	AAG	AAC	ACG	CTC	TAT	CTG	CAA	ATG	AAG	AGC		AGA		- <del>-</del> -	
_		 NI		K	 N		L	Y		0	М	К	s	L	R	Α	E	D
	٠. ك	10								_								224
			279			288			297			306						
• • •	ACG	GCT	GTA	TAT	TAT	TGI	GCG	AGA	GAG	AAG	GCG	CTT	CGG	GGA	ATC	AG I	AGA	TAC
						 C		 R				L		G	I	S	R	Y
	T	А	V	Y	1	Ċ	A	K										
			333	ì		342	2		351			360	)		369	)		
	AA	TAT	TAC	CTO	GAG	GTO	TGG	GGC	AAC	GGC	ACC	ACG	GTC	ACC	GTC	TCC	TCA	3'
					<del>-</del> -									 Т		 S		
	N	Y	Y	۲	D	V	W	G	K	G	T	T	V	Ţ	V	3	ی	
			— CI	DR3 -			<del></del>											

Fig. 16b

#### LD2-20-VL sequence

			9															
5'	GTG	ATG	ACC	CAG	TCT	CCA	TCC	TCC	CTG	TCT	GCA	TCT	GTA	GGA	GAC	AGA	GTC	ACC
					- <b>-</b> -			- <b></b>	- <b></b>									
	V	M	T	Q	S	P	S	S	L	S	A	S	V	G	D	R	V	Т
						72			0 7			9.0			99			108
			63			7.2	CNC	300	ν.α.α. Ο Τ	NCC.	NCC	ייי אייי	ע ידיד	ידממ	TGG	тат	CAG	
	ATC	ACT	TGC	CGG	GCA	AGI	CAG	AGC	AII	AGC	AGC	141						
	 I	т	C	 R	Α	 S	0	s	I	S	S	Y	L	N	W	Y	Q	Q
	-	•	Ŭ	4			_		— ст	ו או				<b></b> →				
			117	-		126									153			162
	מממ	CCA	CCC.	AAA	GCC	CCT	AAG	CTC	CTG	ATC	TAT	GCT	GCA	TCC				
	AAA	CCA																
	K	P	G	K	А	P	К	L	L	I	Y	Α	Α	S	S	L	Q	S
												<del></del>			- CD	R2 —		<b></b> →
			171			180			189			198			207			216
	GGG	GTC	CCA	TCA	AGG	TTC	AGT	GGC	AGT	GGA	TCT	GGG	ACA	GAT	TTC	ACT	CTC	ACC
	G	V	P	S	R	F	S	G	S	G	S	G	T	D	F	T	L	Т
						224			247			252			261			270
			225	CTG	~	234	C	CAT	243	CCN	хст							
	ATC	AGC	AGT	CIG	CAA	CCI	GAA	GAI	111	GCA	ACI	IAC						
		S	s	L	0	P	E	D	F	А	Т	Y	Y	С	0	0	S	Y
٠.	Ι	5	3	1.1	V			D	•	••	-	-	_	_	_			
•			279			288			297			306			315	-		
• :	AGT	ACC	CGA	TTC	ACT	TTC	GGC	CCT	GGG	ACC	AAA				AAA	3 '		
					<b>-</b>									<b>-</b>				
	s	T	R	F	Τ	F	G	P	G	T	K	V	D	I	K			
		(	CDR3		<del></del>													

Fig. 17a

#### LD1-6-17-VH sequence

5'	CAG	GTG	9 AAA	CTG	CTC	GAG	TCT	GGG	27 GGA		GTG	36 GTC	CAG	CCT	45 GGG	AGG	TCC	54 CTG
	Q	v	K	L	L	E	s	G			v	v	Q.	P	G	R	S	L
	AGA	CTT	63 TCC	TGT	GCA	72 GCG	TCT	GGA	81 TTT	ACC	TTC	90 AGT	AGC	TAT	99 GGA	ATG	CAC	108 TGG
	 R	 L	S	 C	Α	<b>-</b> А	s	G	F	Т	F	s	S	Y	G CDR1	M	н	W
	GTC	CGC	117 CAG	GCT	CCA	126 GGA		GGG	135 CTG	GAG	TGG	144 GTG	ACA		153			162 GAT
	v	R	Q	A	P	G	K	G		E	W	V	T	D	I	W	F	D
	GGA	GGT	171 AAT	AAA	CAT	180 TAT	GCA	GAC	189 TTC	GTG	AAG	198 GGC		TTC	207			216
	G	G	N	K	H	Y			F	V	K	G	R	F	Т	I	S	R
	- GAC	TAA	225 TCC	AAG		234	R2 — GGG		243 CTA	CAA	ATG	252 AAC	AGC	CTG	261 AGA		GAG	270 GAC
	·	N	S	к	 N	T	G	F	- <b>-</b> -	Q	М	N	S	L	R	V	E .	D
. 2	ACG	GCT	279 GTC	TAT	TAC	288 TGT	. GCG	AGG	297 GAT	TAC	TAT	306 AGC	GTT	ACT	315 AAG		CTC	324 AGA
	 T	Α	v,	Y	Y	С	A	R	D			S			_	K	L	R
	CT	CA(	331 TAC	3 C TAC	C TAC	342 TAC	: C ATC	GAC	351 351	1		360 AAA			369		C ACC	378 GTC
	L	Н	Y		Y TDR3	Y	M	D	V	· · · · · · · · · · · · · · · · ·	G	K	G	Т	Т	V	Т	V

TCC TCA 3

• 9 9

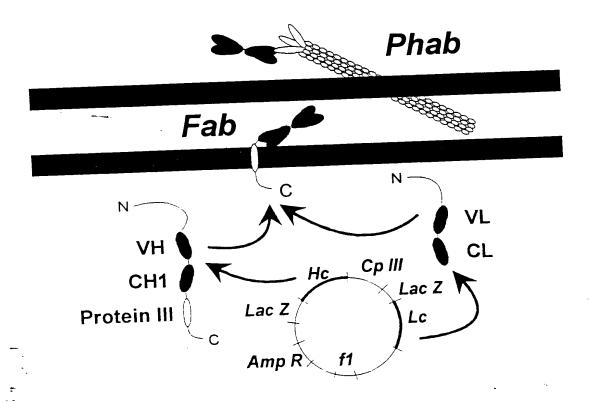
# Fig. 17b

# LD1-6-17-VL sequence

5'	GTG A	ATG	9 ACC	CAG	TCT	18 CCA	TCC	TCC	27 CTG	TCT	GCA	36 TCT	GTA	GGA	GAC	AGA	GTC	ACC
5	v	 М	 T	0	 S	 P		 S	 L		A	S	V	G	D	R	V	Т
	•		_	_	GCA	72 AGT	CAG						TTA		TGG	TAT	CAG 	108 CAA 
	 I	T	C	R	A	S		G				D	L	T 	₩ >	Y	Q	Q
	<b>,</b> , , ,	CCA	117	← AAA :	GCC	126 CCT		CTC				144 GCT	GCA	TCC	153 AAT	TTA	CAA	162 AGT
		 P			 A		 K	-	 L			A			N	L	Q	5
	•		_		a ago	180 TTC	) AGC	: GGC	189 AGT	GG#	A TCT	198 r GG	B C AC	A GA	207	,		216 ACC
					 R									ם	F	Т	L	Т
_												25 T TA	2 T TA	C TG	26: T CT.	1 A CA 	A GA'	270 r AAC
					 J Q				 F						L	Q	D	N
: - 2.5	-				AC AC				29 G GG	7 G AC	C AA	3.0 AG CT	)6 rg G/	AG AT	3·1 C AA	5 \A 3		
	 N	 F	·	P .		· F	·						ւ 1	Ε :	I F	(		

Fig. 18

# The pComb3 Expression System



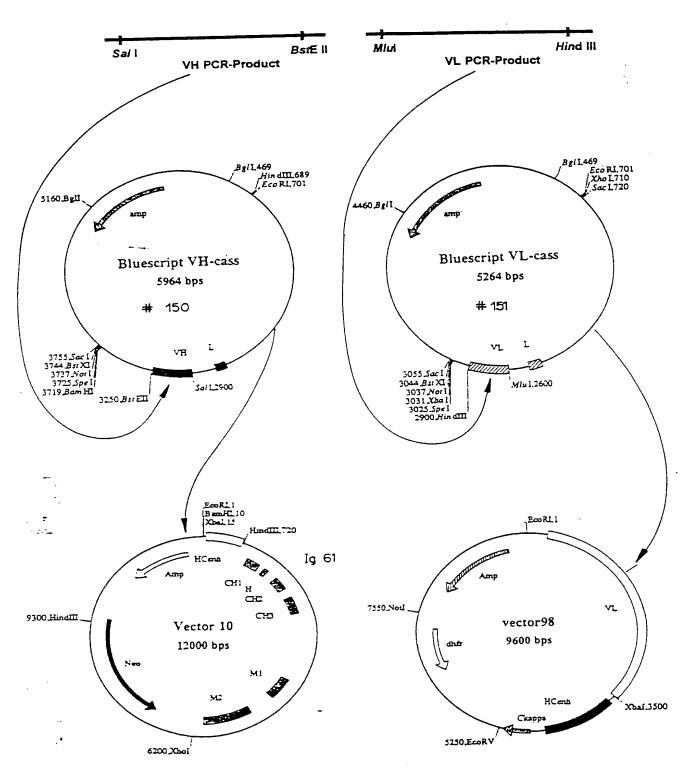


Fig. 19

Fig. 20

